NOVEL PROTEINS AND NUCLEIC ACIDS ENCODING SAME

RELATED APPLICATIONS

This application claims priority from Provisional Applications U.S.S.N. 60/228191, filed August 25, 2000, U.S.S.N. 60/267300, filed February 8, 2001, U.S.S.N. 60/269961, filed February 20, 2001, and U.S.S.N. 60/277337, filed March 20, 2001, each of which is incorporated by reference in its entirety.

FIELD OF THE INVENTION

The invention generally relates to novel nucleic acids and polypeptides encoded thereby.

BACKGROUND OF THE INVENTION

Eukaryotic cells are subdivided by membranes into multiple functionally distinct compartments that are referred to as organelles. Each organelle includes proteins essential for its proper function. These proteins can include sequence motifs often referred to as sorting signals. The sorting signals can aid in targeting the proteins to their appropriate cellular organelle. In addition, sorting signals can direct some proteins to be exported, or secreted, from the cell.

One type of sorting signal is a signal sequence, which is also referred to as a signal peptide or leader sequence. The signal sequence is present as an amino-terminal extension on a newly synthesized polypeptide chain. A signal sequence can target proteins to an intracellular organelle called the endoplasmic reticulum ("ER").

The signal sequence takes part in an array of protein-protein and protein-lipid interactions that result in translocation of a polypeptide containing the signal sequence through a channel in the ER. After translocation, a membrane-bound enzyme, named a signal peptidase, liberates the mature protein from the signal sequence.

The ER functions to separate membrane-bound proteins and secreted proteins from proteins that remain in the cytoplasm. Once targeted to the ER, both secreted and membrane-bound proteins can be further distributed to another cellular organelle called the Golgi apparatus. The Golgi directs the proteins to other cellular organelles such as vesicles, lysosomes, the plasma membrane, mitochondria and microbodies.

Secreted and membrane-bound proteins are involved in many biologically diverse activities. Examples of known secreted proteins include human insulin, interferon, interleukins, transforming GENX-beta, human growth hormone, erythropoietin, and lymphokines. Only a limited number of genes encoding human membrane-bound and secreted proteins have been identified.

The invention generally relates to nucleic acids and polypeptides encoded by them. More specifically, the invention relates to nucleic acids encoding cytoplasmic, nuclear, membrane bound, and secreted polypeptides, as well as vectors, host cells, antibodies, and recombinant methods for producing these nucleic acids and polypeptides.

SUMMARY OF THE INVENTION

The invention is based in part upon the discovery of nucleic acid sequences encoding novel polypeptides. The novel nucleic acids and polypeptides are referred to herein as NOVX, or NOV1, NOV2, NOV3, NOV4, NOV5, NOV6, NOV7, NOV8, NOV9, NOV10, NOV11, NOV12, NOV13, NOV14, NOV15 and NOV16 nucleic acids and polypeptides. These nucleic acids and polypeptides, as well as variants, derivatives, homologs, analogs and fragments thereof, will hereinafter be collectively designated as "NOVX" nucleic acid or polypeptide sequences.

In one aspect, the invention provides an isolated NOVX nucleic acid molecule encoding a NOVX polypeptide that includes a nucleic acid sequence that has identity to the nucleic acids disclosed in SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102. In some embodiments, the NOVX nucleic acid molecule will hybridize under stringent conditions to a nucleic acid sequence complementary to a nucleic acid molecule that includes a protein-coding sequence of a NOVX nucleic acid sequence. The invention also includes an isolated nucleic acid that encodes a NOVX polypeptide, or a fragment, homolog, analog or derivative thereof. For example, the nucleic acid can encode a polypeptide at least 80% identical to a polypeptide comprising the amino acid sequences of SEQ ID NOS:1, 8, 10, 12, 18, 20, 26, 28, 34, 36, 42, 44, 50, 52, 54, 60, 62, 64, 70, 72, 74, 76, 82, 89, 91, 99 and 101. The nucleic acid can be, for example, a genomic DNA fragment or a cDNA molecule that includes the nucleic acid sequence of any of SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102.

Also included in the invention is an oligonucleotide, *e.g.*, an oligonucleotide which includes at least 6 contiguous nucleotides of a NOVX nucleic acid (*e.g.*, SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102) or a complement of said oligonucleotide.

Also included in the invention are substantially purified NOVX polypeptides (SEQ ID NOS:1, 8, 10, 12, 18, 20, 26, 28, 34, 36, 42, 44, 50, 52, 54, 60, 62, 64, 70, 72, 74, 76, 82, 89, 91, 99 and 101). In certain embodiments, the NOVX polypeptides include an amino acid sequence that is substantially identical to the amino acid sequence of a human NOVX polypeptide.

The invention also features antibodies that immunoselectively bind to NOVX polypeptides, or fragments, homologs, analogs or derivatives thereof.

In another aspect, the invention includes pharmaceutical compositions that include therapeutically- or prophylactically-effective amounts of a therapeutic and a pharmaceutically-acceptable carrier. The therapeutic can be, *e.g.*, a NOVX nucleic acid, a NOVX polypeptide, or an antibody specific for a NOVX polypeptide. In a further aspect, the invention includes, in one or more containers, a therapeutically- or prophylactically-effective amount of this pharmaceutical composition.

In a further aspect, the invention includes a method of producing a polypeptide by culturing a cell that includes a NOVX nucleic acid, under conditions allowing for expression of the NOVX polypeptide encoded by the DNA. If desired, the NOVX polypeptide can then be recovered.

In another aspect, the invention includes a method of detecting the presence of a NOVX polypeptide in a sample. In the method, a sample is contacted with a compound that selectively binds to the polypeptide under conditions allowing for formation of a complex between the polypeptide and the compound. The complex is detected, if present, thereby identifying the NOVX polypeptide within the sample.

The invention also includes methods to identify specific cell or tissue types based on their expression of a NOVX.

Also included in the invention is a method of detecting the presence of a NOVX nucleic acid molecule in a sample by contacting the sample with a NOVX nucleic acid probe or primer, and detecting whether the nucleic acid probe or primer bound to a NOVX nucleic acid molecule in the sample.

In a further aspect, the invention provides a method for modulating the activity of a NOVX polypeptide by contacting a cell sample that includes the NOVX polypeptide with a compound that binds to the NOVX polypeptide in an amount sufficient to modulate the activity of said polypeptide. The compound can be, *e.g.*, a small molecule, such as a nucleic acid, peptide, polypeptide, peptidomimetic, carbohydrate, lipid or other organic (carbon containing) or inorganic molecule, as further described herein.

Also within the scope of the invention is the use of a therapeutic in the manufacture of a medicament for treating or preventing disorders or syndromes including, *e.g.*, those described for the individual NOVX nucleotides and polypeptides herein, and/or other pathologies and disorders of the like.

The therapeutic can be, *e g.*, a NOVX nucleic acid, a NOVX polypeptide, or a NOVX-specific antibody, or biologically-active derivatives or fragments thereof.

For example, the compositions of the present invention will have efficacy for treatment of patients suffering from the diseases and disorders disclosed above and/or other pathologies and disorders of the like. The polypeptides can be used as immunogens to produce antibodies specific for the invention, and as vaccines. They can also be used to screen for potential agonist and antagonist compounds. For example, a cDNA encoding NOVX may be useful in gene therapy, and NOVX may be useful when administered to a subject in need thereof. By way of non-limiting example, the compositions of the present invention will have efficacy for treatment of patients suffering from the diseases and disorders disclosed above and/or other pathologies and disorders of the like.

The invention further includes a method for screening for a modulator of disorders or syndromes including, *e.g.*, the diseases and disorders disclosed above and/or other pathologies and disorders of the like. The method includes contacting a test compound with a NOVX polypeptide and determining if the test compound binds to said NOVX polypeptide. Binding of the test compound to the NOVX polypeptide indicates the test compound is a modulator of activity, or of latency or predisposition to the aforementioned disorders or syndromes.

Also within the scope of the invention is a method for screening for a modulator of activity, or of latency or predisposition to an disorders or syndromes including, *e.g.*, the diseases and disorders disclosed above and/or other pathologies and disorders of the like by administering a test compound to a test animal at increased risk for the aforementioned disorders or syndromes. The test animal expresses a recombinant polypeptide encoded by a NOVX nucleic acid. Expression or activity of NOVX polypeptide is then measured in the test animal, as is expression or activity of the protein in a control animal which recombinantly-expresses NOVX polypeptide and is not at increased risk for the disorder or syndrome. Next, the expression of NOVX polypeptide in both the test animal and the control animal is compared. A change in the activity of NOVX polypeptide in the test animal relative to the control animal indicates the test compound is a modulator of latency of the disorder or syndrome.

In yet another aspect, the invention includes a method for determining the presence of or predisposition to a disease associated with altered levels of a NOVX polypeptide, a NOVX nucleic acid, or both, in a subject (*e.g.*, a human subject). The method includes measuring the amount of the NOVX polypeptide in a test sample from the subject and comparing the amount of the polypeptide in the test sample to the amount of the NOVX polypeptide present in a control sample. An alteration in the level of the NOVX polypeptide in the test sample as compared to the control sample indicates the presence of or predisposition to a disease in the subject. Preferably, the predisposition includes, *e.g.*, the diseases and disorders disclosed above and/or other pathologies and disorders of the like. Also, the expression levels of the new polypeptides of

the invention can be used in a method to screen for various cancers as well as to determine the stage of cancers.

In a further aspect, the invention includes a method of treating or preventing a pathological condition associated with a disorder in a mammal by administering to the subject a NOVX polypeptide, a NOVX nucleic acid, or a NOVX-specific antibody to a subject (*e.g.*, a human subject), in an amount sufficient to alleviate or prevent the pathological condition. In preferred embodiments, the disorder, includes, *e.g.*, the diseases and disorders disclosed above and/or other pathologies and disorders of the like.

In yet another aspect, the invention can be used in a method to identity the cellular receptors and downstream effectors of the invention by any one of a number of techniques commonly employed in the art. These include but are not limited to the two-hybrid system, affinity purification, co-precipitation with antibodies or other specific-interacting molecules.

Unless otherwise defined, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this invention belongs. Although methods and materials similar or equivalent to those described herein can be used in the practice or testing of the present invention, suitable methods and materials are described below. All publications, patent applications, patents, and other references mentioned herein are incorporated by reference in their entirety. In the case of conflict, the present specification, including definitions, will control. In addition, the materials, methods, and examples are illustrative only and not intended to be limiting.

Other features and advantages of the invention will be apparent from the following detailed description and claims.

DETAILED DESCRIPTION OF THE INVENTION

The present invention provides novel nucleotides and polypeptides encoded thereby. Included in the invention are the novel nucleic acid sequences and their polypeptides. The sequences are collectively referred to as "NOVX nucleic acids" or "NOVX polynucleotides" and the corresponding encoded polypeptides are referred to as "NOVX polypeptides" or "NOVX proteins." Unless indicated otherwise, "NOVX" is meant to refer to any of the novel sequences disclosed herein. Table A provides a summary of the NOVX nucleic acids and their encoded polypeptides.

TABLE A. Sequences and Corresponding SEQ ID Numbers

NOVX	Internal Identification	SEQ ID NO (nt)	SEQ ID NO (aa)	Homology
NOV1	24CS017	1	2	Kinesin like protein; Overlaps genomic clone with KIAA1236-like protein, predicted secreted
NOV2	24CS059; CG56403-01; 146556340	8	9	Novel Nuclear Protein -like protein
NOV3	24SC113; CG56383-01	10, 12	11	LIM-domain-containing Prickle-like, secreted -like protein
NOV4	24SC128; CG56824-01; 13374351; 13374350; 13374349	18, 20	19	hypothetical protein similar to Y71F9B.2 PROTEIN - Caenorhabditis elegans-like protein
NOV5	24SC239; 13374166; 13374167; 13374355; 13374356; 13374357; 13374358; 13374359; 13374360; 13374361; 13374362	26, 28	27	CG8441 PROTEIN-like protein
NOV6	24SC300	34, 36	35	eEIF-2B epsilon subunit-like protein
NOV7	24SC526; 13374363; 13374364; 13374365; 13374366	42, 44	43	heat shock factor binding protein 1-like protein
NOV8	24SC714; 13373973; 13373974	50	51	putative secreted protein-like protein
NOV9	6CS060; 13374352; 13374353; 13374354	52, 54	53	Kelch-like protein-like protein
NOV10	100340173; 1373975; 1373976; 1373977; 1373978	60, 62, 64	61, 63, 65	hypothetical 22.2 kDa protein SLR0305-like protein; Transmembrane
NOV11	87938450;	70	71	transposase-like protein
NOV12	87917235; 13373979; CG92002-01	72	73	Novel Leucine Zipper Containing Type II membrane like protein-like protein
NOV13	87919652;	74, 76	75	P07948 tyrosine-protein kinase LYN-like protein
NOV14	87935554;	82	83	O15438 canalicular multispecific organic anion transporter 2-like protein; multidrug resistance
NOV15a	100399281	89	90	novel intracellular thrombospondin domain containing protein-like protein
NOV15b	CG57356-01; 159518754	91	92	novel intracellular thrombospondin domain containing protein-like protein
NOV16a	101330077	99	100	FYVE finger-containing phosphoinositide kinase-like protein
NOV16b	CG57248-01; 100391903	101	102	FYVE finger-containing phosphoinositide kinase-like protein

NOVX nucleic acids and their encoded polypeptides are useful in a variety of applications and contexts. The various NOVX nucleic acids and polypeptides according to the invention are useful as novel members of the protein families according to the presence of

domains and sequence relatedness to previously described proteins. Additionally, NOVX nucleic acids and polypeptides can also be used to identify proteins that are members of the family to which the NOVX polypeptides belong.

The NOVX genes and their corresponding encoded proteins are useful for preventing, treating or ameliorating medical conditions, *e.g.*, by protein or gene therapy. Pathological conditions can be diagnosed by determining the amount of the new protein in a sample or by determining the presence of mutations in the new genes. Specific uses are described for each of the sixteen genes, based on the tissues in which they are most highly expressed. Uses include developing products for the diagnosis or treatment of a variety of diseases and disorders.

For example, NOV1 is homologous to a kinesin-like superfamily of proteins. Thus, the NOV1 nucleic acids, polypeptides, antibodies and related compounds according to the invention will be useful in therapeutic and diagnostic applications implicated in, for example; cancer (*e.g.* renal and/or gastric cancer), neurodegenerative diseases, diseases of vesicular transport, and infectious diseases. and/or other pathologies, diseases and disorders.

Also, NOV2 is homologous to the Novel Nuclear Protein-like family of proteins. Thus NOV2 nucleic acids, polypeptides, antibodies and related compounds according to the invention will be useful in therapeutic and diagnostic applications implicated in, for example; cancer and/or other pathologies, diseases and disorders.

Further, NOV3 is homologous to a family of LIM-domain-containing Prickle-like proteins. Thus, the NOV3 nucleic acids and polypeptides, antibodies and related compounds according to the invention will be useful in therapeutic and diagnostic applications implicated in, for example; dystonia-parkinsonism syndrome; dyskeratosis, hereditary benign intraepithelial; developmental disorders, diseases of cytoskeletal function, cancer (*e.g.* gastric, uterine, lung and/or renal cancer), neurodegenerative diseases (*e.g.* Alzheimer's disease, multiple sclerosis and stroke) and/or other pathologies, diseases and disorders.

Also, NOV4 is homologous to the hypothetical protein similar to Y71F9B.2 PROTEIN - Caenorhabditis elegans-like family of proteins. Thus, NOV4 nucleic acids, polypeptides, antibodies and related compounds according to the invention will be useful in therapeutic and diagnostic applications implicated in, for example; heart disease, stroke, autoimmune disease, infectious disease, and cancer (*e.g.* renal and/or breast cancer) and/or other pathologies, diseases and disorders.

Additionally, NOV5 is homologous to the CG8441 PROTEIN-like family of proteins. Thus NOV5 nucleic acids, polypeptides, antibodies and related compounds according to the invention will be useful in therapeutic and diagnostic applications implicated in, for example; cancer (*e.g.* breast and/or ovarian cancer) and/or other pathologies, diseases and disorders.

Also, NOV6 is homologous to the eEIF-2B epsilon subunit-like family of proteins. Thus NOV6 nucleic acids, polypeptides, antibodies and related compounds according to the invention will be useful in therapeutic and diagnostic applications implicated in, for example; cancer (*e.g.* breast and/or ovarian cancer) and/or other pathologies, diseases and disorders.

Further, NOV7 is homologous to members of the heat shock factor binding protein 1-like family of proteins. Thus, the NOV7 nucleic acids, polypeptides, antibodies and related compounds according to the invention will be useful in therapeutic and diagnostic applications implicated in, for example; cancer (*e.g.* breast and/or ovarian cancer) and/or other pathologies, diseases and disorders.

Still further, NOV8 is homologous to the putative secreted protein-like protein family of proteins. Thus, NOV8 nucleic acids and polypeptides, antibodies and related compounds according to the invention will be useful in therapeutic and diagnostic applications implicated in, for example; cancer (*e.g.* liver, lung, ovarian and/or colon cancer), inflammatory diseases and/or other pathologies, diseases and disorders.

Additionally, NOV9 is homologous to the Kelch-like protein-like family of proteins. Thus, NOV9 nucleic acids and polypeptides, antibodies and related compounds according to the invention will be useful in therapeutic and diagnostic applications implicated in Menkes disease, myoglobinuria/hemolysis due to PGK deficiency, and Wieacker-Wolff syndrome, neurological disorders, development-related pathologies and/or other various pathologies, diseases and disorders.

NOV10a, NOV10b and NOV10c are homologous to a hypothetical 22.2 kDa protein SLR0305-like protein family of proteins and the Type IIIb plasma membrane-like family of proteins. Thus, the NOV10 nucleic acids, polypeptides, antibodies and related compounds according to the invention will be useful in therapeutic and diagnostic applications implicated in, for example; ACTH deficiency; Convulsions, familial febrile, 1; Duane syndrome; congenital Adrenal hyperplasia due to 11-beta-hydroxylase deficiency; glucocorticoid-remediable Aldosteronism; congenital Hypoaldosteronism due to CMO I deficiency; congenital Hypoaldosteronism due to CMO II deficiency; susceptibility to Nijmegen breakage syndrome; Low renin hypertension; Anemia, Ataxia-telangiectasia, Autoimmune disease, Immunodeficiencies, kidney cancer, proliferative disease, immune-mediated disease, allergy, asthma, and psoriasis and/or other pathologies, diseases and disorders.

NOV11 is homologous to a transposase-like protein family of proteins. Thus, the NOV11 nucleic acids, polypeptides, antibodies and related compounds according to the invention will be useful in, for example; potential therapeutic applications such as the following: (i) a protein therapeutic, (ii) a small molecule drug target, (iii) an antibody target (therapeutic,

diagnostic, drug targeting/cytotoxic antibody), (iv) a nucleic acid useful in gene therapy (gene delivery/gene ablation), (v) an agent promoting tissue regeneration *in vitro* and *in vivo*, and (vi) a biological defense weapon, and/or transposase-related pathologies, diseases and disorders.

Also, NOV12 is homologous to the Novel Leucine Zipper Containing Type II membrane like protein-like family of proteins. Thus NOV12 nucleic acids, polypeptides, antibodies and related compounds according to the invention will be useful in therapeutic and diagnostic applications implicated in, for example; prostate cancer, lung cancer, diabetes, abnormal wound healing, congenital slow-channel myosthenic syndrome, asthma, IBD, contact hypersensitivity, infection disease, allorejection, autoimmunity, inflammation and/or other pathologies, diseases and disorders.

Further, NOV13 is homologous to a family of P07948 tyrosine-protein kinase LYN-like proteins. Thus, the NOV13 nucleic acids and polypeptides, antibodies and related compounds according to the invention will be useful in therapeutic and diagnostic applications implicated in, for example; breast cancer, diabetes and/or other pathologies, diseases and disorders.

Also, NOV14 is homologous to the O15438 canalicular multispecific organic anion transporter 2-like family of proteins. Thus, NOV14 nucleic acids, polypeptides, antibodies and related compounds according to the invention will be useful in therapeutic and diagnostic applications implicated in, for example: detoxification, drug resistance, multidrug resistance, inflammatory disease, cancer, liver disease and/or other pathologies, diseases and disorders.

Additionally, NOV15 is homologous to the novel intracellular thrombospondin domain containing protein-like family of proteins. Thus NOV15 nucleic acids, polypeptides, antibodies and related compounds according to the invention will be useful in therapeutic and diagnostic applications implicated in, for example; systemic lupus erythematosus, autoimmune disease, asthma, emphysema, scleroderma, allergy, ARDS; fertility, breast cancer, liver differentiation, hypogonadism; angiogenesis, vascularization in CNS tissue undergoing repair/regeneration, CNS-related cancers, diseases of the thyroid gland, immunological disease, diseases of the thyroid gland and pancreas as well as other metabolic and neuroendocrine diseases and/or other pathologies, diseases and disorders.

Also, NOV16a and NOV16b are homologous to the FYVE finger-containing phosphoinositide kinase-like family of proteins. Thus NOV16 nucleic acids, polypeptides, antibodies and related compounds according to the invention will be useful in therapeutic and diagnostic applications implicated in, for example; diabetes, obesity, fertility, signaling and/or other pathologies, diseases and disorders.

The NOVX nucleic acids and polypeptides can also be used to screen for molecules, which inhibit or enhance NOVX activity or function. Specifically, the nucleic acids and

polypeptides according to the invention may be used as targets for the identification of small molecules that modulate or inhibit, *e.g.*, neurogenesis, cell differentiation, cell proliferation, hematopoiesis, wound healing and angiogenesis.

In one embodiment of the present invention, NOVX or a fragment or derivative thereof may be administered to a subject to treat or prevent a disorder associated with decreased expression or activity of NOVX. Examples of such disorders include, but are not limited to, cancers such as adenocarcinoma, leukemia, lymphoma, melanoma, myeloma, sarcoma, teratocarcinoma, and, in particular, cancers of the adrenal gland, bladder, bone, bone marrow, brain, breast, cervix, gall bladder, ganglia, gastrointestinal tract, heart, kidney, liver, lung, muscle, ovary, pancreas, parathyroid, penis, prostate, salivary glands, skin, spleen, testis, thymus, thyroid, and uterus; neurological disorders such as epilepsy, ischemic cerebrovascular disease, stroke, cerebral neoplasms, Alzheimer's disease, Pick's disease, Huntington's disease, dementia, Parkinson's disease and other extrapyramidal disorders, amyotrophic lateral sclerosis and other motor neuron disorders, progressive neural muscular atrophy, retinitis pigmentosa, hereditary ataxias, multiple sclerosis and other demyelinating diseases, bacterial and viral meningitis, brain abscess, subdural empyema, epidural abscess, suppurative intracranial thrombophlebitis, myelitis and radiculitis, viral central nervous system disease, prion diseases including kuru, Creutzfeldt-Jakob disease, and Gerstmann-Straussler-Scheinker syndrome, fatal familial insomnia, nutritional and metabolic diseases of the nervous system, neurofibromatosis, tuberous sclerosis, cerebelloretinal hemangioblastomatosis, encephalotrigeminal syndrome, mental retardation and other developmental disorders of the central nervous system, cerebral palsy, neuroskeletal disorders, autonomic nervous system disorders, cranial nerve disorders, spinal cord diseases, muscular dystrophy and other neuromuscular disorders, peripheral nervous system disorders, dermatomyositis and polymyositis, inherited, metabolic, endocrine, and toxic myopathies, myasthenia gravis, periodic paralysis, mental disorders including mood, anxiety, and schizophrenic disorders, akathesia, amnesia, catatonia, diabetic neuropathy, tardive dyskinesia, dystonias, paranoid psychoses, postherpetic neuralgia, and Tourette's disorder; and disorders of vesicular transport such as cystic fibrosis, glucose-galactose malabsorption syndrome, hypercholesterolemia, diabetes mellitus, diabetes insipidus, hyper- and hypoglycemia, Grave's disease, goiter, Cushing's disease, Addison's disease, gastrointestinal disorders including ulcerative colitis, gastric and duodenal ulcers, other conditions associated with abnormal vesicle trafficking including acquired immunodeficiency syndrome (AIDS), allergic reactions, autoimmune hemolytic anemia, proliferative glomerulonephritis, inflammatory bowel disease, multiple sclerosis, myasthenia gravis, rheumatoid arthritis, osteoarthritis, scleroderma, Chediak-Higashi syndrome, Sjogren's syndrome, systemic lupus erythiematosus, toxic shock syndrome,

traumatic tissue damage, and viral, bacterial, fungal, helminthic, and protozoal infections, as well as additional indications listed for the individual NOVX clones.

The NOVX nucleic acids and proteins of the invention are useful in potential diagnostic and therapeutic applications and as a research tool. These include serving as a specific or selective nucleic acid or protein diagnostic and/or prognostic marker, wherein the presence or amount of the nucleic acid or the protein are to be assessed. These also include potential therapeutic applications such as the following: (i) a protein therapeutic, (ii) a small molecule drug target, (iii) an antibody target (therapeutic, diagnostic, drug targeting/cytotoxic antibody), (iv) a nucleic acid useful in gene therapy (gene delivery/gene ablation), (v) an agent promoting tissue regeneration *in vitro* and *in vivo*, and (vi) a biological defense weapon.

Additional utilities for the NOVX nucleic acids and polypeptides according to the invention are disclosed herein.

NOV1

A disclosed NOV1 nucleic acid of 1065 nucleotides (also referred to as 24CS017) encoding a novel kinesin-like protein is shown in Table 1A. An open reading frame was identified beginning with an ATG initiation codon at nucleotides 1-3 and ending with a TAA codon at nucleotides 1063-1065. The start and stop codons are shown in bold letters in Table 1A.

Table 1A. NOV1 nucleotide sequence (SEQ ID NO:1).

ATGACGGGGCTGCTCCTCCTCAGCCTCCAGTCAGGCTGTGTGGCAGCGATCACCTCCATGTCGATGGAGTGTCTGTG
CAGTTTGGGAGCGAGGCTCTGCCTCTCTCGGTCTACCCTTGGGAGTGAAATAGTGACCGTCCCTTTGAGCCCGAGAG
CTGGGGAGAAGGCCGTGCCTGTTAACAGCTGCCTGGACCCTCTCTGGAGAGCAGCAGAGAGAGGCGGGGCTGAGAGA
GATGTTGCCAAGAACCTAAGGGTGAAAGTCATGCTTCGCATCTGTTCCACCTTGGCTCGAGATACTTCAGAATCCAG
CTCTTTCTTAAAGGTGGACCCACGGAAGAAGCAGATCACCTTGTACGATCCCCTGACTTGTGGAGGTCAAAATGCCT
TCCAAAAGAGAGGCAACCAGGTTCCTCCAAAGATGTTTGCCTTCGATGCAGTTTTTCCACAAGACGCTTCTCAGGCT
GAAGTGTGTGCAGGCACCGTGGCAGAGGTGATCCAGTCTGTGGTCAACGGGGCAGATGGCTGCGTGTTCTGTTTCGG
CCACGCCAAACTGGGAAAATCCTACACCATCATCGAAAGGATGATTCCATGCAGAACCTGGCATCATTCCCTGTG
CCATCTCTTGGCTCTTCAAGCTCATAAACGAACGCAAGGAAAAGACCGGCGCCCGTTTCTCAGTCCGGGTTTCCGCC
GTGGAAGTGTGGGGGAAGGAGGAGAACCTGCGGGACCTGCTGTCGGAGGTGGCCACGGGCAGCCTGCAGGACGGCCA
GTCCCCGGGCGTGTACCTCTGTGAGGACCCCATCTGCGGCACGCAGCTGCAGAACCAGAGCGAGCTGCGGGCCCCCA
CCGCAGAGAAGGCTGCCTTTTTCCTGGATGCCGCCATTGCCTCCCGCAGGAGCCACCAACAGGACTGTGATGAGGAC
GACCACCGCAACTCACACGTGTTCTTCACACTGCACATCTACCAGTACCGGATGGAGAAGAGCGGGAAAGGGGGAAT
TCTGCTTTCGATTTGGAATCTGAAAGTAGGGAGAAATCTTGAAAACAAGGAAACAGTTCA**TTAA**

A disclosed NOV1 polypeptide (SEQ ID NO:2) encoded by SEQ ID NO:1 has 354 amino acid residues and is presented in Table 1B using the one-letter amino acid code. SignalP, Psort and/or Hydropathy results predict that NOV1 has a signal peptide and is likely to be localized extracellularly with a certainty of 0.4562. In an alternative embodiment, NOV1 is likely to be localized to the endoplasmic reticulum membrane with a certainty of 0.1000, or to the endoplastic reticulum lumen with a certainty of 0.1000, or to the microbody (peroxisome)

with a certainty of 0.1000. The most likely cleavage site for a NOV1 peptide is between amino acids 16 and 17, *i.e.*, at the dash between amino acids VAA-IT. NOV1 has a molecular weight of 38525.7 Daltons.

Table 1B. Encoded NOV1 protein sequence (SEQ ID NO:2).

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MTGLLLLSLQSGCVAA/ITSMSMECLCSLGARLCLSRSTLGSEIVTVPLSPRAGEKAVPVNSCLDPLWRAAERGGAGGD
VAKNLRVKVMLRICSTLARDTSESSSFLKVDPRKKQITLYDPLTCGGQNAFQKRGNQVPPKMFAFDAVFPQDASQAEVC
AGTVASVIQSVVNGADGCVFCFGHAKLGKSYTMIGKDDSMQNLGIIPCAISWLFKLINERKEKTGARFSVRVSAVEVWG
KEENLRDLLSEVATGSLQDGQSPGVVLCEDPICGTQLQNQSELRAPTAEKAAFFLDAAIASRRSHQQDCDEDDHRNSHV
FFTLHIYQYRMEKSGKGGILLSIWNLKVGRNLENKETVH
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In all BLAST alignments herein, the "E-value" or "Expect" value is a numeric indication of the probability that the aligned sequences could have achieved their similarity to the BLAST query sequence by chance alone, within the database that was searched. The Expect value (E) is a parameter that describes the number of hits one can "expect" to see just by chance when searching a database of a particular size. It decreases exponentially with the Score (S) that is assigned to a match between two sequences. Essentially, the E value describes the random background noise that exists for matches between sequences.

The Expect value is used as a convenient way to create a significance threshold for reporting results. The default value used for blasting is typically set to 0.0001. In BLAST 2.0, the Expect value is also used instead of the P value (probability) to report the significance of matches. For example, an E value of one assigned to a hit can be interpreted as meaning that in a database of the current size one might expect to see one match with a similar score simply by chance. An E value of zero means that one would not expect to see any matches with a similar score simply by chance. See, *e.g.*, http://www.ncbi.nlm.nih.gov/Education/BLASTinfo/.

In a search of public sequence databases, NOV1 was found to have homology to the amino acid sequences shown in the BLASTP data listed in Table 1C.

Table 1C. BLASTP results for NOV1

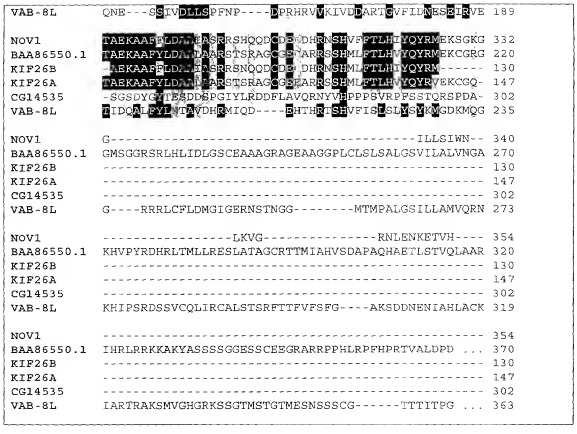
Gene Index/ Identifier	Protein/ Organism	Length (aa)	Identity (%)	Positives (%)	Expect
Q9ULI4; AB033062; BAA86550.1	KIAA1236 PROTEIN (FRAGMENT) homo sapiens 6/2001	1481	155/222 (70%)	185/222 (83%)	4e-87
Q99PU2; KIF26B; BAB32487	KINESIN SUPERFAMILY PROTEIN 26B (FRAGMENT). KIF26B, mus musculus 6/2001	130	122/145 (84%)	126/145, (87%)	7e-64
Q99PT4; AB054031; BAB32495.1;	KINESIN SUPERFAMILY PROTEIN 26A (FRAGMENT). KIF26A, mus musculus 6/2001	147	106/147 (72%)	130/147, (88%)	2e-58
Q9VLW2; AE003619; AAF52569.1	CG14535 PROTEIN. drosophila melanogaster 6/2001	302	69/165 (42%)	99/165, (60%)	9e-28

Q9U541; AF108229; AAF17300.1	VAB-8L. caenorhabditis elegans 6/2001	1066	61/191 (32%)	98/191, (51%)	1e-18

The homology of these and other sequences is shown graphically in the ClustalW analysis shown in Table 1D. In the ClustalW alignment of the NOV1 protein, as well as all other ClustalW analyses herein, the black outlined amino acid residues indicate regions of conserved sequence (*i.e.*, regions that may be required to preserve structural or functional properties), whereas non-highlighted amino acid residues are less conserved and can potentially be mutated to a much broader extent without altering protein structure or function.

Table 1D. ClustalW Analysis of NOV1

```
1)  Novel NOV1  (SEQ ID NO:2)
2)  BAA86550.1  partial sequence used (SEQ ID NO:3)
3)  KIF26B  (SEQ ID NO:4)
4)  KIF26A (SEQ ID NO:5)
5)  CG14535  (SEQ ID NO:6)
6)  VAB-8L - partial sequence used (SEQ ID NO:7)

NOV1          MTGLLLLSLQSGCVAAITSMSMECLCSLGARLCLSRSTLGSEIVTVPLSP 50
BAA86550.1    -------------------------------------------------- 1
KIF26B        -------------------------------------------------- 1
KIF26A        -------------------------------------------------- 1
CG14535       ----------------------------------------MATTSTSNMS 10
VAB-8L        -------------------------------------------------- 1

NOV1          RAGEKAVPVNSCLDPLWRAABRGGAGGDVAKNLRVKVMLRICSTLARDTS 100
BAA86550.1    -------------------------------------------------- 1
KIF26B        -------------------------------------------------- 1
KIF26A        -------------------------------------------------- 1
CG14535       RNGGFCGALQRAPPPMPPTLIRRLSSRECYGVGKVKVMLRVADRDRNSGG 60
VAB-8L        ----------------------------------------MEACSSKTSLL 11

NOV1          ESSSFLKVDPRKKQITTYDP-LTCGGQNAPQKRGNQVP--PKMFAFTAVE 147
BAA86550.1    -------------QVILYDP-AAGPPGSAGPRRAATAAV-PKMFAPTAVE 35
KIF26B        -------------------------------------------------- 1
KIF26A        -------------------------------------------------- 1
CG14535       TEPDFMALDKKRQVTLTDPRTACPPPQAAQERAPMVAA-PKMFAFTNLE 109
VAB-8L        LHSPLRTIPKLRLCASISSEDVAHGRCSLTDQHLQIEGKNYSKTTPQHIE 61

NOV1          PQDASQAEVCAGTVAEVTQSWVNGADGCVFCFGHAKLGKSYTMIGGLDSM 197
BAA86550.1    PQDSPQAEYCSGTVADVLQSWVSGADGCIFSFGHMSLGKSYTMIGGSSP 85
KIF26B        ----------------------------------PAPTGKSYTMIGGLDSM 17
KIF26A        ----------------------------------GKSYTMIGGSSP 13
CG14535       TGEDKQSDVCASALSSVTPAVLEGDGCLLAMGYPATGAQTWLGSLGGG 159
VAB-8L        RTDATQDDMYTAFLSDTINSVPAQDATVLAMGAKTNGKDERDYGNSVER 111

NOV1          Q-----------NLGIIPCAISWLFLLIERKEKTCARFSVRVSAVEVW 235
BAA86550.1    Q-----------SLGIIPCAISWLFLLIERKEKTCARFSVRVSAVEVC 123
KIF26B        Q-----------NLGIIPCAISWLFLLIERKEKTCARFSVRISAVEVW 55
KIF26A        Q-----------SLGIIPCAISWLFLLIERKEKTCARFSFRVSAVEVC 51
CG14535       SGSGSASGSGVACSLCASIAWIYKGLIERLQKSCARFSVRVSAVEIS 209
VAB-8L        N-----------SLVQMAIDQIMNALDDNKD-SPERIQVRVSAIMIS 146

NOV1          GKE---MVLRDLLSEVAQGSLQDGSPGVYLQEDPVCGTQLQNQSPRRAP 282
BAA86550.1    GRD---ASLRDLLAEVAQGSLQDNSPGVYLQSDPVCAQLQNQSPRRAP 170
KIF26B        GKE---PNLRDLLSEVAQGSLQDGLSPGVYLQEDPVCGTQLQNQSPRRAP 87
KIF26A        GHD---ASLRDLLAEVAQGSLQDTSPGVYLQEDPVCGTQLQNQSPRRAP 98
CG14535       ATKPDAISQDLLISHLVEYGVYSHIKPNALFIHSGLLFWSSVWN----- 254
```



Other BLAST results include sequences from the Patp database, which is a proprietary database that contains sequences published in patents and patent publications. Patp results include those listed in Table 1E.

Table 1E. Patp BLASTP Analysis for NOV1

Sequences producing High-scoring Segment Pairs	Protein/ Organism	Length (aa)	Identity (%)	Positive (%)	E Value
patp:AAY51328	Human KLIMP protein-*H. sapiens*	1103	29	48	1.6e-11
Patp:AAB36227	Human kinesin-like protein HKLP	1816	29	49	8.2e-11
patp:AAB94768	Human protein SEQ ID NO:15849-*H. sapiens*	664	29	50	6.3e-10
Patp:AAY06618	Thermomyces lanuginosus Kinesin motor protein TL-gamma-*Thermomyces lanuginosus*	784	26	46	1.4e-09
Patp:AAY01632	Amino acid sequence of centromere-associated protein-E - *Xenopus sp*	2954	38	56	1.8e-08
Patp:AAG21666	Arabidopsis thaliana protein fragment SEQ ID NO: 24303 - *Arabidopsis thaliana*	452	30	57	2.7e-08

The presence of identifiable domains in NOV1, as well as all other NOVX proteins, was determined by searches using software algorithms such as PROSITE, DOMAIN, Blocks, Pfam, ProDomain, and Prints, and then determining the Interpro number by crossing the domain match (or numbers) using the Interpro website (http:www.ebi.ac.uk/ interpro). DOMAIN results for NOV1 as disclosed in Tables 1F, were collected from the Conserved Domain Database (CDD) with Reverse Position Specific BLAST analyses. This BLAST analysis software samples domains found in the Smart and Pfam collections.

Table 1F lists the domain description from DOMAIN analysis results against NOV1. This indicates that the NOV1 sequence has properties similar to those of other proteins known to contain these domains. In a sequence alignment herein, fully conserved single residues are calculated to determine percent homology, and conserved and "strong" semi-conserved residues are calculated to determine percent positives. The "strong" group of conserved amino acid residues may be any one of the following groups of amino acids: STA, NEQK, NHQK, NDEQ, QHRK, MILV, MILF, HY, FYW.

Table 1F. Domain Analysis of NOV1

```
Prodom analysis
Sequences producing High-scoring Segment Pairs.                  High    Smallest Sum
                                                                 Score   Probability
                                                                         P(N)
prdm:361   p36 (52) KINH(7) KINN(2) KF1(2)  // PROTEIN M...     189     3.2e-15
prdm:12025 p36 (2) CYT1(2)  // PROBABLE B-TYPE CYTOCHROME...     55     0.76
prdm:29378 p36 (1) RPSW_STRCO // RNA POLYMERASE SIGMA FAC...     57     0.93
prdm:14019 p36 (2) CIK6(2)  // CHANNEL VOLTAGE-GATED POTA...     49     0.998
prdm:44434 p36 (1) ERY1_SACER // ERYTHRONOLIDE SYNTHASE, ...     49     0.998

>prdm:361 p36 (52) KINH(7) KINN(2) KF1(2) // PROTEIN MOTOR ATP-BINDING
MICROTUBULES COILED COIL KINESIN-LIKE CELL KINESIN MITOSIS, 170 aa.
 Identities = 43/108 (39%), Positives = 66/108 (61%)
 for NOV1: 139 to 246, and Sbjct: 61 to 168

>prdm:12025 p36 (2) CYT1(2) // PROBABLE B-TYPE CYTOCHROME TRICARBOXYLIC ACID CYCLE
ELECTRON TRANSPORT HEME TRANSMEMBRANE, 48 aa.
 Identities = 13/21 (61%), Positives = 15/21 (71%)

>prdm:29378 p36 (1) RPSW_STRCO // RNA POLYMERASE SIGMA FACTOR WHIG. TRANSCRIPTION
REGULATION; SIGMA FACTOR; DNA-DIRECTED RNA POLYMERASE; DNA-BINDING, 81 aa.
 Identities = 14/42 (33%), Positives = 21/42 (50%)

>prdm:14019 p36 (2) CIK6(2) // CHANNEL VOLTAGE-GATED POTASSIUM PROTEIN KV1.6 IONIC
TRANSMEMBRANE ION TRANSPORT GLYCOPROTEIN, 40 aa.
 Identities = 9/19 (47%), Positives = 13/19 (68%)

>prdm:44434 p36 (1) ERY1_SACER // ERYTHRONOLIDE SYNTHASE, MODULES 1 AND 2 (EC
2.3.1.94) (ORF 1) (6- DEOXYERYTHRONOLIDE B SYNTHASE I) (DEBS 1). TRANSFERASE;
ACYLTRANSFERASE; ANTIBIOTIC BIOSYNTHESIS; NADP; PHOSPHOPANTETHEINE;
MULTIFUNCTIONAL ENZYME, 55 aa.
Identities = 14/35 (40%), Positives = 16/35 (45%)

BLOCKS analysis
AC#        Description                                          Strength  Score
BL00411C   Kinesin motor domain proteins.                         1642   1283
BL00411B   Kinesin motor domain proteins.                         1185   1156
BL00411D   Kinesin motor domain proteins.                         1217   1107
BL00853G   Beta-eliminating lyases pyridoxal-phosphate a         1858   1105
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BL00509B  Ras GTPase-activating proteins.                    1280   1073
BL01227A  Uncharacterized protein family UPF0012 protei      1059   1072
BL00094F  C-5 cytosine-specific DNA methylases proteins      1186   1045
BL01240B  Purine and other phosphorylases family 2 prot      1350   1039
BL00487G  IMP dehydrogenase / GMP reductase proteins.        1525   1029
BL00411A  Kinesin motor domain proteins.                     1284   1019
BL00370B  PEP-utilizing enzymes phosphorylation site pr      1554   1015
BL00838C  Interleukins -4 and -13 proteins.                  1661   1011
BL00486A  DNA mismatch repair proteins mutS family prot      1290   1010

ProSite analysis                                        NOV1 aa position
Pattern-ID: ASN_GLYCOSYLATION PS00001 (Interpro)                     275
Pattern-DE: N-glycosylation site, Pattern: N[^P][ST][^P]

Pattern-ID: GLYCOSAMINOGLYCAN PS00002 (Interpro)                     329
Pattern-DE: Glycosaminoglycan attachment site, Pattern: SG.G


Pattern-ID: PKC_PHOSPHO_SITE PS00005 (Interpro)          49, 226, 297, 329
Pattern-DE: Protein kinase C phosphorylation site
Pattern: [ST].[RK]


Pattern-ID: CK2_PHOSPHO_SITE PS00006 (Interpro)      61, 116, 152, 160, 230, 252
Pattern-DE: Casein kinase II phosphorylation site
Pattern: [ST].{2}[DE]

Pattern-ID: MYRISTYL PS00008 (Interpro)                 12, 29, 73, 124, 171,
Pattern-DE: N-myristoylation site                          201, 222, 256, 333
Pattern: G[^EDRKHPFYW].{2}[STAGCN][^P]

Pattern-ID: ATP_GTP_A PS00017 (Interpro)                             180
Pattern-DE: ATP/GTP-binding site motif A (P-loop)
Pattern: [AG].{4}GK[ST]
```

The disclosed NOV1 nucleic acid encoding a kinesin-like protein includes the nucleic acid whose sequence is provided in Table 1A, or a fragment thereof. The invention also includes a mutant or variant nucleic acid any of whose bases may be changed from the corresponding base shown in Table 1A while still encoding a protein that maintains its kinesin -like activities and physiological functions, or a fragment of such a nucleic acid. The invention further includes nucleic acids whose sequences are complementary to those just described, including nucleic acid fragments that are complementary to any of the nucleic acids just described. The invention additionally includes nucleic acids or nucleic acid fragments, or complements thereto, whose structures include chemical modifications. Such modifications include, by way of nonlimiting example, modified bases, and nucleic acids whose sugar phosphate backbones are modified or derivatized. These modifications are carried out at least in part to enhance the chemical stability of the modified nucleic acid, such that they may be used, for example, as antisense binding nucleic acids in therapeutic applications in a subject. In the mutant or variant nucleic acids, and their complements, up to about 60% percent of the bases may be so changed.

The disclosed NOV1 protein of the invention includes the kinesin -like protein whose sequence is provided in Table 1B. The invention also includes a mutant or variant protein any of

whose residues may be changed from the corresponding residue shown in Table 1B while still encoding a protein that maintains its kinesin-like activities and physiological functions, or a functional fragment thereof. In the mutant or variant protein, up to about 60 % percent of the residues may be so changed.

The invention further encompasses antibodies and antibody fragments, such as F_{ab} or $(F_{ab})_2$, that bind immunospecifically to any of the proteins of the invention. Also encompassed within the invention are peptides and polypeptides comprising sequences having high binding affinity for any of the proteins of the invention, including such peptides and polypeptides that are fused to any carrier partcle (or biologically expressed on the surface of a carrier) such as a bacteriophage particle.

Kinesin family proteins are microtubule-based motor proteins that drive the transport of molecular component within the cell. Translocation of components within the cell is critical for maintaining cell structure and function.

Kinesin defines a ubiquitous, conserved family of over 50 proteins that can be classified into at least 8 subfamilies based on primary amtino acid sequence, domain structure, velocity of movement, and cellular function. See review in: Moore and Endow (1996) Bioessays 18:207-219; and Hoyt (1994) Curr. Opin. Cell Biol. 6:63-68). The prototypical kinesin molecule is involved in the transport of membrane-bound vesicles amd organelles. This function is particularly important for axonal transport in neurons. Protein-containing vesicles are constantly transported from the neuronal cell body along microtubules that span the length of the axon leading to the synaptic terminal. Failure to supply the synaptic terminal with these vesicles blocks the transmission of neural signals. In the fruit fly Drosophila melanogaster, for example, mutations in kinesin cause severe disruption of axonal transport in larval nerves which leads to progressive paralysis. See Hurd and Saxton (1996) Genetics 144:1075-1085. This phenotype mimics the pathology of some vertebrate motor neuron diseases, such as amyotrophic lateral sclerosis (ALS). In addition to axonal transport, kinesin is also important in all cell types for the transport of vesicles from the Golgi complex to the endoplasmic reticulum. This role is critical for maintaining the identity and functionality of these secretory organelles.

Members of the more divergent subfamilies of kinesin are called kinesin-related proteins (KRPs), many of which function during mitosis in eukaryotes as divergent as yeast and human (Hoyt, *supra*). Some KRPs are required for assembly of the mitotic spindle. *In vivo* and *in vitro* analyses suggest that these KRPs exert force on microtubules that comprise the mitotic spindle, resulting in the separation of spindle poles. Phosphorylation of KRP is required for this activity. Failure to assemble the mitotic spindle results in abortive mitosis and chromosomal aneuploidy, the latter condition being characteristic of cancer cells. In addition, a unique KRP, centromere

protein E. localizes to the kinetochore of human mitotic chromosomes and may play a role in their segregation to opposite spindle poles.

As described earlier, NOV1 shares extensive sequence homologies with kinesin family proteins, including kinesin superfamily protein 26A and 26B, and with kinesin-like proteins, including human kinesin-like motor protein (KLIMP), human kinesin-like protein (HKLP) and Thermomyces lanuginosus Kinesin motor protein TL-gamma. The structural similarities indicate that NOV1 may function as a member of kinesin family proteins. Therefore, NOV1, like kinesin family proteins and kinesin-related proteins, may be associated with cancer, neurological disorders and disorders of vehicular transport. Accordingly, the NOV1 nucleic acids and proteins identified here may be useful in potential therapeutic applications implicated in (but not limited to) various pathologies and disorders as indicated herein. For example, a cDNA encoding the kinesin-like protein NOV1 may be useful in gene therapy, and the kinesin-like protein NOV1 may be useful when administered to a subject in need thereof. The NOV1 nucleic acid encoding kinesin-like protein, and the kinesin-like protein of the invention, or fragments thereof, may further be useful in diagnostic applications, wherein the presence or amount of the nucleic acid or the protein are to be assessed. Additional disease indications and tissue expression for NOV1 is presented in Example 2.

Based on the tissues in which NOV1 is most highly expressed, specific uses include developing products for the diagnosis or treatment of a variety of diseases and disorders.

NOV1 nucleic acids and polypeptides are further useful in the generation of antibodies that bind immuno-specifically to the novel NOV1 substances for use in therapeutic or diagnostic methods. These antibodies may be generated according to methods known in the art, using prediction from hydrophobicity charts, as described in the "Anti-NOVX Antibodies" section below. The disclosed NOV1 protein has multiple hydrophilic regions, each of which can be used as an immunogen. In one embodiment, a contemplated NOV1 epitope is from about amino acids 50 to 80. In another embodiment, a NOV1 epitope is from about amino acids 100 to 150. In additional embodiments, NOV1 epitopes are from about amino acids 190 to 200, from about amino acids 205 to 275 and from about amino acids 280 to 330. These novel proteins can be used in assay systems for functional analysis of various human disorders, which will help in understanding of pathology of the disease and development of new drug targets for various disorders.

NOV2

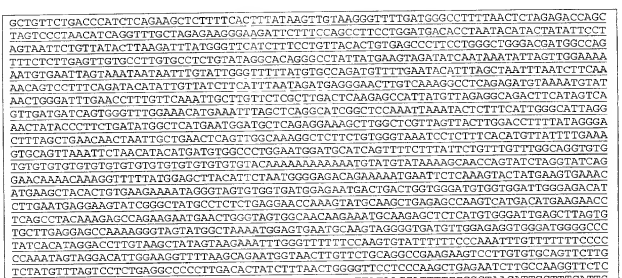
A disclosed NOV2 nucleic acid of 7560 nucleotides (also referred to as 24CS059, CG56403-01 and 146556340) encoding a novel nuclear protein-like protein is shown in Table

2A. An open reading frame was identified beginning with an ATG initiation codon at nucleotides 7170-7172 and ending with a TGA codon at nucleotides 7476-7478. A putative untranslated region upstream from the initiation codon and downstream from the termination codon is underlined in Table 2A, and the start and stop codons are in bold letters.

5

Table 2A. NOV2 nucleotide sequence (SEQ ID NO:8).

GTATTCTCAGAGCTGCCAGGAGTGCATCGAGCCTGTAATTTCCTGTTCTCTGAACTCCCCATCTTTCTGCAGCTCCAAGCTT
TGTGTCCCACAGCCTTGTGACTCTGTGCTAACAAATCGCTATTGTCCAGTGGGGCGAATGGTGGCTGGAACTAAAGAATTGCT
GTCTGGTTTCTATTCAAATCCAGGTAGCGAGATAATAGAATGGACTTTTCGAATCGTCATGTGAATAACGTCTGCTCGGCAT
GAAGGCTCAGAGCCATGCTAGGAAGGATTAACTCGTAGGCTGACCACTAACATCCTTTGTGGTACGAGGGAGAAACATTCCC
AAGTATCATTTTATTCACACTTAATTTTCTATCCCATACCCCCAAAATAAGGCTAGCTATTAATTAGTTGGCTGCTTTTCT
CTTAATTTTTAGTGTTTCTGTTGATAATGTGTAAGTTTGGGAAAATGCTAAGTAGCTTTTCACTTAGAACACTGTTATTTTC
TCTTTAAAGTTTTCTACCTTACATTTATTATAGCATAGTATCTTTTATAGCATAGATGCAGAAAGTAAGAGAGAGCTTGTTT
TTTCAAGAAAACAACCCTTTAAAATACTTTCCAACCCATGAAGGGAAAAATCCTCCTTTTTTCCCCCAAGTGCATTCTACTT
ATTACTTTGCATTTTTCTCCCAAAGTCCAAATTTATGCAAAGAAAATAGAAACAAGTTCAAATGCAATGCATTAACCAAATA
AAACAAGTCTGCTTCAAATTAGGAACCAACCTAAGCATTTGTAAAGTGTAGCAGAATCAGAATTCTTTTAAAAATTAGATTT
GGAACCTGAACTATATAATTCTCATTTTTCTGTGGAAAATTATTTTATCTTTCTCTGTATACCTGAAAAAATGT
CCATAGGCTTAAAGGGTCATGCTTTTACATTCCTTCCATATCACAGGTACTATGAAGTAAGGAGACTTTTAGGTTCCTTTTT
GTCTTAAACTCAGACAGCTTTGTAAGCAGTAGTGTGTAGATTACAAGAGTTAGACAAAAGCAGGCGCGACTGAGAAGAGTTG
GTGGGGGAGAAGCTTGGGGCACTTCCTGTCACTCAACACATTCCAGATCACTAAAAAATTTCCAGACCCTCTCAGTTCCCCC
TTGCCCACTCCAGTTCCGGTTATTTCTGATTCCATATGTTGTGGTATTTACCATACTTCTCTCTCCTCACTAGGCTCTGGCA
AGACTGCTTCAGAGGGGATGCATTCCTTTAGATTGCACAAAGCGGAGCTGGGAAAATGGCTGGCAGTTTCAGAATCTAGTCA
CGATCGCACGCATGAGCACCTCACACATCCATGTCCCTACCCGCCCCCCGCTCCGCCCCTGCAGCTGGCTGACCTGTCTC
ACCCACTGCTGGCCTATCGAACCGGCAGGACTGTCTGGTTTGGCTCGTGCCTTTGTCCATGTCTGGCTTAGTTCCTCTCTG
CTATGCTTGCCTCTACCCCCACCGCCCCAGGCGGCACAAGTGTTGGCCACACAAAACTAGAGATAGAAAAGGTGGTAAAA
ACTTCAAACTTTCTAAATTCTCCAACAGTTTATTTCTTGTGAATTTCTTCCTCTTTAAATACTCCATTTTAAGAAAACAA
AAAAATTAATTATCTAAAGGCAAAGAATGGAAAGCAACCTTTGTGTTCCTTATAATAACTGACTTCATAACTCTCTCCAGCT
GCGTTATGGGATGTGTATAAAAAGCTTCTGTTCTGAGAACAAAGGAGCACGTGCAGAAATGAGACGAAAAATCCACTGACA
GTATTCCATTACACAAATTACTTAAAAGATTTTAGTCAAGCCCCTCAACAGATTCAATTTTAAAATGGCTTTTAGTTAAAAA
AAAAAAATTGAAAGTCTTACCCAGTAAAAGAACCGAAGTAGTCCTGAACTGTTACGTAAGACTTTTTACAGTTGGATCTTT
GTCAAAAGGGGATGGGGGTGATGGGAGAAAGCAGCAACGACAATCAAAAAAGTTCGAGCTGCTGTGGCTAGAGGACAACTTC
TGTGTTTCCAGATAGGATTCTTGCTGTAGAAATGGAACTTCCAGCCAGCACAGCATCCTGTCCCAGTAGAGAAATGAGTTTG
TCAGTTAAAACAAAAAAAAAAAATTAGATACTGGAACCCAGGCTAGACGAGGTATTGAACCGCGCAGATTTCCTTCGCCCCT
GTCTGCTCAGCTCGCATTGAACTATATATGACCCAGATGATGGACAGAAGCACATTAGTCATGTGCACACTGGAAGAAAGC
GGATTTGCTGGTCCCTGGCAGTGCAGGGGTTTGTCTTCTGATTGGCTGTGCCTGATCGGTGAAATGTGAAGCCCTCACCA
TTTCAGAGGCCGTAATTCAGGACTGGCAGTTTGAGTGTCTGGCTGCCTCTAGTCACTGAGAGACTTTGAAGGTGTTGCTTTTG
TTTGGTGGCATTACCCACCCGAGGTTGCTTACACCTCTCTACTTGTGTCAGAAGAAATACTAGTCTTTCTGAAATACAAAT
AGGCAGCCGATTTTCCTCTGAATCCTAAATCACCCTATTGTTGATAAACTTGGCTCTAACTGAAACCAATTATTTGATTTGAA
AATTTATTGTGATCCTAACCAAGCTTCATATCCAGACCAACCCTTGGTCTTGATTTTATAGGTTTGATAAGGTAAAAATAAA
AGTGGCATATTTGACTTTGAAGCCTCTATATGATATAAATTGCTCTTAATGAAAATTGGATAGATGGACAACAGAGAAGTGA
AGTTTTAGATTCTGGAGTGTTTGGATGTATGAGGAAGAAGCTTTATGTCTTTTATCCCCTTTGTCAGACTGTCACTCTTGT
CCCAGTCCTAGTCACATTAGGGGTTGCTGGGGGGGGGAAGCTATGAAAGCATGGACCCTACTGAGCTGTGACATAGCCTTTA
ATCATGCAAGACAGCCACGGTCTGCTCTCTTCAGTCTGTCTGAACTAGGGTCCTTGGGGTTTATTTTCCATCTTTCTGAGCC
ACTGGGAAACCAGGTCATTATACAGGACTGTCATTTGTGACATTTTGTTTAGTACATGGCAGTTGCTTTGTTTATTTAATG
CAAGTTGACACTTCTTTAAAGTTTCAAAACAGTAAAGTTGTTTTGTGAGACCTTGACTCTGATATATGAAATCTACTCTACA
TGGACCAATCATTTTTTTTCCGTGGACTTTCTTGTCTCTTAGAAATTAGCTTATAGAGTCCTAAATTGATACTTAAACATAC
CAATAGTCTGTTTATTTCTTGCCTTTCTCACAGTTGTTGAAATAATTCCATCTGTCTTTTGCTGTAAATTTTGGGTTTG
GATGTTTGTACTTGGAATTTTTTAGATGTTGACTATATTATCAGCACCTTCCATATGAGGACTACCCAGAATTATTCTCT
TGTCTTAACCCGAGAAAAGCTGTTTGATGCACTATTAGATAAGAATGTTCGAAAGAAGAGGAGATGAGCACTCTCTTGC
TTTTTGTAAGCCACAAGAGCAATCTTTTTTTTTCTAAGTTGTGGTAAGGTATATGTAACATAAAATTGACTGTTGTAATAAT
TTTTAAGTGTATAGTTCTGTGGTGTTAAGTGCATTCACGTTGTTTTGCAGCCTTCACCACCATCCATCCACCACAGAACTCT
TCTCCCTTTGCAAAACTGAAATTCTCTACCTACCTGTTAAACACTAACTTGCCATTCTTCCCTCCCCCAGGCCCTGGGGACA
ACCATCATTCTACTTTCTCTTTGATTTTTTGTTTTTTGTTTTTGGAGACGGAATTTTACTCTTGTTGCCCAAGCTGGGATGC
AATGGCACTGTCTTGGCTCACTGCAACCTTCCGCCTCCTGGGTTCAAGCAATTCTCCTTCCTCAGCCTCCTGAGTAGCTGGGA
CTACAGGTGCCCACCACCACGCCTGGCTAGTTTTTGTATTTTTAGTAGACACGGGGTTTCACCATGTTGGCCAGGCTGGTCT
CGAACTCGTGATCTCAAGTGATCCACCCACCTTGGCCTCCCAAAATCCTAGAATTACAGGCATGAGCCACCGTGCCTAGCC
TCTGTCTGTTTGCTTTTTGACTACTCTAGATACCTCATATAAGTGGAATAATACAAGATGTGTTCCCTTTTGACAGGCTTAT
TTCACTTAGCATGGTGCCTCCAAGGTTCATGCATGTTGTCGCATGTCAGAATTTCCTTACGTTTGAAGGCTGAATAATATAC
CATTGCATGTGATGTATACTACTGTCTTAGTCCCTTTAGTGTTGCTGTAAAGGAATACCTGAGGCTGGGTAATTTATAAAGAAAA
GAGGTTTATTTGGCTCATGGTTCTGCAAGCTGTACAGAAGACATGGCACCAGCTTCTGGTGAGGGCCTCAAGCTGCCTCAT
TCATGGCACAAGGTGAAAGGGAGCTGGTGTGTGCAGAGATCACATGGTAGGAGAGGAGGAGGCAAGAGAGAGAGAAGGAGG
TGCCAGACTACTTTAAAACCATCAGCTTTTGCAGGAGTTATAGAGCCAGCACTCACTACTACTGCAAGAATGGCACCAAG
ACATTCATGAGGGATCTGCCTTCATGACCAGAACACCTCCCACCAGGCCCCACCACAATAAGGGGTTAGATTTCAGCAT
GAGACTCAATGAGGGGGAGCAACAAATTACATCCAAACTGTAGCAACCACATTTTGTTTATCCATTCATCTGTCAATGGA
CACTTAAGTAGCTTCCACTTTTTTGCTATCAAGACAGTTTTTCTTGACTATTCTTAAAATCATGTGAGGGCTTCTTTACAGA



The NOV2 nucleic acid was identified on chromosome 9 by comparing the sequence to public databases. The NOV2 nucleic acid maps to the 9q33-34 locus, a region associated with endotoxin hyporesponsiveness (OMIM 603030), adrenocortical insufficiency without ovarian defect (OMIM 184757) and other diseases/disorders. Single nucleotide polymorphisms were identified for NOV2, as described in Example 3. It was found that NOV2 had homology to the nucleic acid sequences shown in the BLASTN data listed in Table 2B.

Table 2B. BLASTN results for NOV2

Gene Index/ Identifier	Protein/ Organism	Begin-End	Length (nt)	Identity (%)	Expect
AL158075	Human DNA sequence from clone RP11-348K2 on chromosome 9q33.1-34.13, complete sequence. 6/2001. Strand = Plus / Minus	[1-7560] [3799-4086] [4584-4654] [5736-5773] [6954-7071] [7003-7071]	102867	7560/7560 (100%)	0.0
AK021895	Homo sapiens cDNA FLJ11833 fis, clone HEMBA1006579. 9/2000.	[1-2237]	2237	2234/2237 (100%)	0.0

BLASTN homology of NOV2 to the GenBank Acc. No. AL158075 genomic clone in Table 2B depicts a proposed exon and intron structure for the NOV2 gene, which is most likely encoded on the AL158075 clone minus strand. The NOV2 nucleic acid is likely to be expressed

in 10 week embryo and whole embryo, mainly head, based on its homology to GenBank Acc. No. AK021895. GenBank AK021895, disclosed in September 2000, has homology to the 5' untranslated NOV2 sequence.

Exons were predicted by homology and the intron/exon boundaries were determined using standard genetic rules, as described in Example 1. Exons were further selected and refined by means of similarity determination using multiple BLAST (for example, tBlastN, BlastX, and BlastN) searches, and, in some instances, GeneScan and Grail. Expressed sequences from both public and proprietary databases were also added when available to further define and complete the gene sequence. The DNA sequence was then manually corrected for apparent inconsistencies thereby obtaining the sequences encoding the full-length protein.

A disclosed NOV2 polypeptide (SEQ ID NO:9) encoded by SEQ ID NO:8 has 102 amino acid residues and is presented in Table 2C using the one-letter amino acid code. SignalP, Psort and/or Hydropathy results predict that NOV2 has no known signal peptide and is likely to be localized in the nucleus with a certainty of 0.300. In alternative embodiments, a NOV2 polypeptide is located in the mitochondrial matrix space with a certainty of 0.100, in a lysosome (lumen) with a certainty of 0.100, or in a microbody (peroxisome) with a certainty of 0.0101. NOV2 has a molecular weight of 11700.6 Daltons.

Table 2C. Encoded NOV2 protein sequence (SEQ ID NO:9).

```
MMMPPYSRMVTETLSLKKQQQNKPLTNTENNSIHLIVPFYRQVTSSIFIVKYHVVSSDTFFFLLKKKSYLQATQLISQLT
EGAARTLESTKGEKTEALVVK
```

No sequences were found in the EMBL, PIR or GenBank databases that had homology to the NOV2 polypeptide in an unfiltered BLASTP search (expectation value=1.0 for input parameter).

The presence of identifiable domains in NOV2, as well as all other NOVX proteins, was determined by searches using software algorithms such as PROSITE, DOMAIN, Blocks, Pfam, ProDomain, and Prints, and then determining the Interpro number by crossing the domain match (or numbers) using the Interpro website (http:www.ebi.ac.uk/ interpro). DOMAIN results for NOV2 as disclosed in Tables 1E, were collected from the Conserved Domain Database (CDD) with Reverse Position Specific BLAST analyses. This BLAST analysis software samples domains found in the Smart and Pfam collections.

Table 2D lists the domain description from DOMAIN analysis results against NOV2. Table 2E provides the percent homologies of NOV2 to the domains found in the BLASP analyses. Homology to one or more domains indicates that the NOV2 sequence has properties similar to those of other proteins known to contain these domains, and is a likely phosphoprotein.

Table 2D. Domain Analysis of NOV2

PRODOM Protein Domain Analysis

Sequences producing High-scoring Segment Pairs:	High Score	Smallest Sum Probability P(N)
prdm:38396 p36 (1) DRTS_PLAFK - DIHYDROFOLATE REDUCTASE ...	51	0.37
prdm:48689 p36 (1) Y360_MYCGE - HYPOTHETICAL PROTEIN MG3...	51	0.37
prdm:55080 p36 (1) DPOM_PODAN - PROBABLE DNA POLYMERASE . .	61	0.69
prdm:16122 p36 (2) PHAC(1) PHBC(1) - POLYMERASE SYNTHAS. .	46	0.84
prdm:24351 p36 (1) RS6_HABIN - 30S RIBOSOMAL PROTEIN S6....	46	0.84

BLOCKS Protein Domain Analysis

AC#	Description	Strength	Score
BL00243G	Integrins beta chain cysteine-rich domain pro	1511	1011
BL00951C	ER lumen protein retaining receptor proteins.	1661	1002
BL01081	Bacterial regulatory proteins, tetR family pr	1354	1002
BL00126A	3'5'-cyclic nucleotide phosphodiesterases pro	1312	1000
BL00764A	Endonuclease III iron-sulfur binding region p	1181	1000

ProSite Protein Domain Analysis — **AA of NOV2 (SEQ ID NO:4)**

Pattern-ID: ASN_GLYCOSYLATION PS00001 (Interpro)
Pattern-DE: N-glycosylation site
Pattern: N[^P][ST][^P] — 30

Pattern-ID: CAMP_PHOSPHO_SITE PS00004 (Interpro)
Pattern-DB: cAMP- and cGMP-dependent protein kinase phosphorylation site
Pattern: [RK]{2}.[ST] — 66

Pattern-ID: PKC_PHOSPHO_SITE PS00005 (Interpro)
Pattern-DE: Protein kinase C phosphorylation site
Pattern: [ST].[RK] — 15, 90

Pattern-ID: CK2_PHOSPHO_SITE PS00006 (Interpro)
Pattern-DE: Casein kinase II phosphorylation site
Pattern: [ST].{2}[DE] — 26, 91

Pattern-ID: MYRISTYL PS00008 (Interpro)
Pattern-DE: N-myristoylation site
Pattern: G[^EDRKHPFYW].{2}[STAGCN][^P] — 83

Table 2E. ProDom results for NOV2

ProDom Identifier	Protein/ Organism	Length (nt)	Identity (%)	Positive (%)	Expect
prdm:38396	p36 (1) DRTS_PLAFK - DIHYDROFOLATE REDUCTASE (EC 1.5.1.3) / THYMIDYLATE SYNTHASE (EC 2.1.1.45) (DHFR-TS). MULTIFUNCTIONAL ENZYME; OXIDOREDUCTASE; TRANSFERASE; NADP; METHYLTRANSFERASE; NUCLEOTIDE BIOSYNTHESIS; ONE-CARBON METABOLISM	52	11/41 (26%)	24/41 (58%)	0.46
prdm:48689	p36 Y360_MYCGE - HYPOTHETICAL PROTEIN MG360	38	14/34 (41%)	19/34 (55%)	0.46
prdm:55080	p36 (1) DPOM_PODAN - PROBABLE DNA POLYMERASE (EC 2.7.7.7) DNA-DIRECTED DNA POLYMERASE	135	14/60 (23%)	28/60 (46%)	1.2
prdm:16122	p36 (2) PHAC(1) PHBC(1) - POLYMERASE SYNTHASE PHA POLY 3-HYDROXYALKANOATE PHA-POLYMERASE POLYHYDROXYALKANOIC ACID BIOSYNTHESIS TRANSFERASE	55	14/37 (37%)	20/37 (54%)	1.8

prdm 24351	36 (1) RS6_HAEIN // 30S RIBOSOMAL PROTEIN S6. RIBOSOMAL PROTEIN; RRNA-BINDING	35	10/23 (43%)	14/23 (60%)	1.8

Other BLAST results include sequences from the Patp database, which is a proprietary database that contains sequences published in patents and patent publications. Patp results include those listed in Table 2F.

Table 2F. Patp alignments of NOV2

PatP Identifier	Protein/ Organism	Length (nt)	Identity (%)	Positive (%)	Expect
AAB43292	Human ORFX ORF3056 polypeptide sequence SEQ ID NO:6112, PN=WO200058473-A2	110	69/101 (68%)	77/101 (76%)	3.4e-29
AAG02872	Human secreted protein, SEQ ID NO: 6953, PN=EP1033401-A2	144	60/101 (59%)	73/101 (72%)	1.1e-25
AAR97079	Respiratory Syncytial Virus antigenic fragment 30	61	15/30 (50%)	17/30 (56%)	2.1
AAR97084	Respiratory Syncytial Virus antigenic fragment 35	51	15/30 (50%)	17/30 (56%)	2.1
AAR97080	Respiratory Syncytial Virus antigenic fragment 31	59	15/30 (50%)	17/30 (56%)	2.1
AAR97081	Respiratory Syncytial Virus antigenic fragment 32	57	15/30 (50%)	17/30 (56%)	2.1
AAR97082	Respiratory Syncytial Virus antigenic fragment 33	55	15/30 (50%)	17/30 (56%)	2.1
AAR97083	Respiratory Syncytial Virus antigenic fragment 34	53	15/30 (50%)	17/30 (56%)	2.1

The disclosed NOV2 nucleic acid encoding a nuclear protein -like protein includes the nucleic acid whose sequence is provided in Table 2A, or a fragment thereof. The invention also includes a mutant or variant nucleic acid any of whose bases may be changed from the corresponding base shown in Table 2A while still encoding a protein that maintains its nuclear protein -like activities and physiological functions, or a fragment of such a nucleic acid. The invention further includes nucleic acids whose sequences are complementary to those just described, including nucleic acid fragments that are complementary to any of the nucleic acids just described. The invention additionally includes nucleic acids or nucleic acid fragments, or complements thereto, whose structures include chemical modifications. Such modifications include, by way of nonlimiting example, modified bases, and nucleic acids whose sugar phosphate backbones are modified or derivatized. These modifications are carried out at least in part to enhance the chemical stability of the modified nucleic acid, such that they may be used, for example, as antisense binding nucleic acids in therapeutic applications in a subject. In the mutant or variant nucleic acids, and their complements, up to about 67% percent of the bases may be so changed.

The disclosed NOV2 protein of the invention includes the nuclear protein -like protein whose sequence is provided in Table 2B. The invention also includes a mutant or variant protein any of whose residues may be changed from the corresponding residue shown in Table 2B while

still encoding a protein that maintains its nuclear protein -like activities and physiological functions, or a functional fragment thereof. In the mutant or variant protein, up to about 66 % percent of the residues may be so changed.

The invention further encompasses antibodies and antibody fragments, such as F_{ab} or $(F_{ab})_2$, that bind immunospecifically to any of the proteins of the invention.

The above defined information for this invention suggests that this nuclear protein -like protein (NOV2) may function as a member of a nuclear protein family. Therefore, the NOV2 nucleic acids and proteins identified here may be useful in potential therapeutic applications implicated in (but not limited to) various pathologies and disorders as indicated herein. The potential therapeutic applications for this invention include, but are not limited to: cancer research tools, for all tissues and cell types composing (but not limited to) those defined here, including cancerous and normal tissue, endotoxin hyporesponsiveness (OMIM 603030), adrenocortical insufficiency without ovarian defect (OMIM 184757) and other diseases/disorders.

The NOV2 nucleic acids and proteins of the invention are useful in potential therapeutic applications implicated in cancer including but not limited to and/or other pathologies and disorders. For example, a cDNA encoding the nuclear protein -like protein (NOV2) may be useful in cancer therapy, and the nuclear protein -like protein (NOV2) may be useful when administered to a subject in need thereof. By way of nonlimiting example, the compositions of the present invention will have efficacy for treatment of patients suffering from diseases including but not limited to endotoxin hyporesponsiveness and cancer. The NOV2 nucleic acid encoding nuclear protein -like protein, and the nuclear protein -like protein of the invention, or fragments thereof, may further be useful in diagnostic applications, wherein the presence or amount of the nucleic acid or the protein are to be assessed.

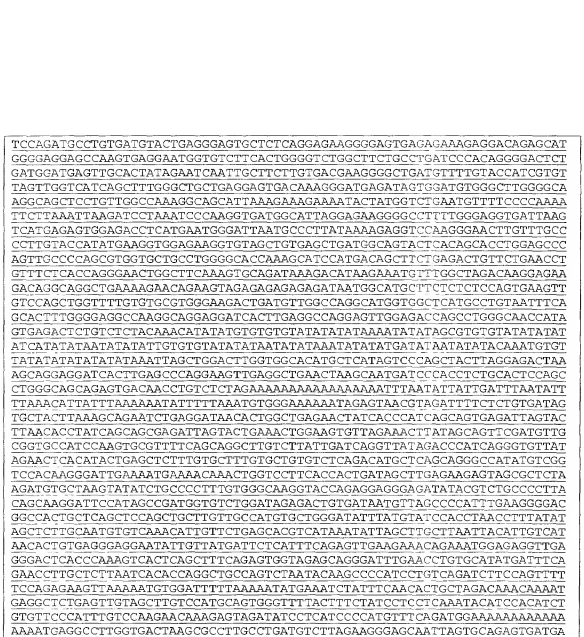
NOV2 nucleic acids and polypeptides are further useful in the generation of antibodies that bind immuno-specifically to the novel NOV2 substances for use in therapeutic or diagnostic methods. These antibodies may be generated according to methods known in the art, using prediction from hydrophobicity charts, as described in the "Anti-NOVX Antibodies" section below. The disclosed NOV2 protein has multiple hydrophilic regions, each of which can be used as an immunogen. In one embodiment, a contemplated NOV2 epitope is from about amino acids 10 to 38. In another embodiment, a NOV2 epitope is from about amino acids 55 to 102. These novel proteins can be used in assay systems for functional analysis of various human disorders, which will help in understanding of pathology of the disease and development of new drug targets for various disorders.

NOV3

A disclosed NOV3 nucleic acid of 7380 nucleotides (also referred to as 24SC113) encoding a novel LIM-domain containing Prickle-like protein is shown in Table 3A. An open reading frame was identified beginning with an ATG initiation codon at nucleotides 1991 to 1993 and ending with a TGA codon at nucleotides 2951 to 2953. The start and stop codons are in bold letters in Table 3A.

Table 3A. NOV3 nucleotide sequence (SEQ ID NO:10).

GTGAGTCAGGGAGGAGAAAGGTAGGCTGCTTGGGCCGGTGGCCTTTTGTTCTTGCAATTCTCTTCTTCTC
CCTAATTTCTGGTTCATTGCCTCTTTAGACAAGTCTCCAGAAGTTCTTCCTTGAAAGTCCAGGCTCAGGA
ACTCTCAGCCACTGAAGATAAAGGCCACATTAGTCCCTTTTTCTGGGAAGCCGTGTATCATTACGCATCA
GGAGAATGCAGGGGTCCTGGTCCACCCTACAGTCATAGCTTGAGGCTATATTCCCAGCAGGCTCTCCCCA
CGGGAAGGGGCCCCAGCAGCTCCCAGTTTCCATTCTGCCAGTTTTACTGCTGCTATAAAAAGAGCCTGCT
GTGTGACTGCCTTAGCAAAAGTCCTGCCTTAGAAAAAGCAATGAGAGGTGTTGGCTTAGTGCAGGTCACT
TGCCCACCCCTGAATCAGTCCCTGGGTGCCAGGAGAGCAGATTTTTTTTGCTGGCCTATGTTGGGCCCCA
GATCAGCTTTTGCCCCACCCAAAGCTCACGGCCTGAAGATGGCAGGGAAATGGTGTCCCACAGGGAGAGG
AAGTCCTATAACCAGAAGAGGGCAGAGATGATGAGAAGGCAGAACCCCTGGGGCTGTGGGAGGCTCCCTT
AGTACGCAGTGTGGCCAGGCTATATAAACCTGGCGCAGGCCTGTCACAGGGAGGAATCGTACCTCTTCCT
TCCCTGATGAAATTAAGCAAAGGGTACTTACGCTCCCAGAGGGGCAGTAGCTTTGGCAATACCGTGTCTA
GGTTTTTCTTTACCGAAAGCAGATTTTTCCTTAACAAGAGTTGAAATCCACATTTTTATTTCCCACTAAG
TCTGTTGAGACTGCTTTAACGGAATAGCACAGACTGGGTGGCCTCTGAGTAACAGAAATGTATTGCTGAC
AGTTCTGAAAGCTGGGAAGTTCAAACTCAAGGCACCAGCAAATGCAGTGTCTGCTGAGGGCCTGTTTTTT
GTTTCCTGGATGATACTTTCTGGCAGAGTCATCATATAGTGGAAGGAGCAAACAGGCTCCCTTGGGCCTC
TGTTATAAGGGCACTAATCTCATTCATGAGGTATCCACTCTCATGACCTAGTCACCTCCCAAAAAGCTCC
ATCTCCTAATGCCATCACTTTAGGATTTAGGTGTTAAACTTAGGAGTTCTGAAGAAAACATTCACCATAG
CATCCACTGAGTTGCTGCTGTGACTTACCCATTGGAATAGCATATGCTAGTAATGGGATTCACTCGATCT
ATCTACACACAAAGAGCCCTGTCATACACCAGGCCATGTTCCAGGTCCTGGAGATGCTGTAGAAACTCAA
TGAGTCTGTCCTCATAGAGCTTCACTTTTAGCGGGGGAGAGAAATAATAAACAGATGCATGTATATACTG
TTGTAATGTAAAGCGGTATTAATGCTATCAAGAAAACTCCAGCAGGTAAGGGTGGAGAGTAATGGAGAAT
CACTATTTAGTGTGGATAGGAAGAACTTCTCAGAGGAGTTGGCTTTGAGCAGATGCCTAACTAGAGTGAA
GGAGATAGTGTCAATGTCATGGTTGAGAATAAGACTTCCTGGGTACAGATCTCGTCTCTGGTTCCTAGTT
ATGTTACCCTGCCAAGTTACTTAGCCTCATCTGCCTCTACTTTCTCATGTGAAAACTGCAAATAATATTA
GAAAGCTAGCTCAAGGAGCTGAGTGATTAAATGAGTTTACATATATAAAGCTCTTAAAGCAGTACATGAT
CATACGTTAATATTACTATTGCTATTTGTCAGGGGGAAATGTGTCCCAGGCACAAGGATTCATAGACAAG
CCATTTTAACCTAGAGTCTTTGTGCTTGGAGCAAATGAGTTAAGGCGCATACTGGTACAACAAGGACTTC
TCGTAATAGGACGTGAATACCATTTACATAAGGGTCTGATTGTTGATTTATTGACAGTTTATCCTGCCGC
ACCTGGAATCCTGAGACAAACCAAGGTGCTATGTGTTTCACGTCCCAGTGCAGAGCTCTGAGCAGCTCAT
CAGCCTCTCCAATGTCTCTCATTTTTTTAGGTATCGACCAAGGTCAAATGACCTATGATGGCCAACACTG
GCATGCACTGAGACCTGTTTCTGCTGTGCTCACTGCAAGAAATCCCTCCTGGGGCGGCCATTCCTCCCG
AAGCACGGCCAGATATTCTGCTCACGGGCCTGCAGTGCTGGGGAAGACCCCAATGGTTCTGACTCCTCTG
ATTCCGCCTTCCAGAACGCCAGGGCCAAGGAGTCCCGGCGCAGTGCCAAAATTGGCAAGAACAAGGGCAA
GACGGAGGAGCCCATGCTGAACCAGCACAGCCAGCTGCAAGTGAGTTCTAACCGGCTGTCAGCCGACGTA
GACCCCCTGTCACTGCAGATGGACATGCTCAGCCTGTCCAGCCAGACACCCAGCCTCAACCGGGACCCCA
TCTGGAGGAGCCGGGAAGAGCCCTACCATTATGGGAACAAGATGGAGCAGAACCAGACCCAGAGCCCTCT
GCAGCTCCTCAGCCAGTGCAACATCAGAACTTCCTACAGTCCAGGAGGGCAAGGGGCTGGGGCCCAGCCC
GAAATGTGGGGCAAGCACTTCAGCAACCCCAAAAGGAGCTCGTCACTGGCCATCACAGGACATGCTGGCA
GCTTCATCAAGGAATGCCGAGAAGACTATTACCCGGGGAGGCTGAGATCTCAGGAGAGCTACAGTGATAT
GTCTAGTCAGAGTTTCAGTGAGACCCGAGGCAGCATCCAAGTCCCCAAATATGAGGAGGAAGAGGAAGAG
GAAGGGGGCTTGTCCACTCAGCAGTGTCGGACCCGTCATCCCATCAGTTCCCTGAAATACACAGAGGACA
TGACGCCCACAGAGCAGACCCCTCGGGGCTCCATGGAATCCCTGGCCCTGTCTAATGCAACAGGTAGGTT
CTGTTCACCTTGAAAACAGATAGAAAGGGGGTAGTCTCTGGGTGACTGGATGCTGGTCCCCAGGAATTTT
TTTTTTTTTTGAAATGGAGTCTCGCTCTGTCCCCAGGCTGGAGTGCAGTGGCACGATCTCCGCTCACTG
CAAGCTCCACCTCCCGGGTTCACGCCATTCTCCTGCCTCAGCCTCACGAGTAGTTGGGACTACAGGTGCC
CGCCACCATGCCTGGCTAATTTTTTTGTATTTTTAGTACACACGTGTTTCACCGTGTTAGCCGGGATGTT
CTCGATCTCCTGACCTCGTGATCCACCTGCCTCGGCCTCCCAAAGTGGTAGGATTACAGGCGTGAGCCAC
CGTGCCCAGCCTGGTCCTCCGGATTTTAATGTTGTTTCTGCCACGTGCCCTCTTCTAATAGGCTGCTGAG
GAAGGTAAACCCAAGTTTGAGATGGCTTCTATCTTTGATGGGCTTCCCTGTAAACAAAGCCTGAGACAGG



TCCAGATGCCTGTGATGTACTGAGGGAGTGCTCTCAGGAGAAGGGGAGTGAGAGAAAGAGGACAGAGCAT
GGGGAGGAGCCAAGTGAGGAATGGTGTCTTCACTGGGGTCTGGCTTCTGCCTGATCCCACAGGGGACTCT
GATGGATGAGTTGCACTATAGAATCAATTGCTTCTTGTGACGAAGGGGCTGATGTTTTGTACCATCGTGT
TAGTTGGTCATCAGCTTTGGGCTGCTGAGGAGTGACAAAGGGATGAGATAGTGGATGTGGGCTTGGGGCA
AGGCAGCTCCTGTTGGCCAAAGGCAGCATTAAAGAAAGAAAATACTATGGTCTGAATGTTTCCCCAAAA
TTCTTAAATTAAGATCCTAAATCCCAAGGTGATGGCATTAGGAGAAGGGGCCTTTTGGGAGGTGATTAAG
TCATGAGAGTGGAGACCTCATGAATGGGATTAATGCCCTTATAAAAGAGGTCCAAGGGAACTTGTTTGCC
CCTTGTACCATATGAAGGTGGAGAAGGTGTAGCTGTGAGCTGATGGCAGTACTCACAGCACCTGGAGCCC
AGTTGCCCCAGCGTGGTGCTGCCTGGGGCACCAAAGCATCCATGACAGCTTCTGAGACTGTTCTGAACCT
GTTTCTCACCAGGGAACTGGCTTCAAAGTGCAGATAAAGACATAAGAAATGTTTGGCTAGACAAGGAGAA
GACAGGCAGGCTGAAAAGAACAGAAGTAGAGAGAGAGAGATAATGGCATGCTTCTCTCTCCAGTGAAGTT
GTCCAGCTGGTTTTGTGTGCGTGGGAAGACTGATGTTGGCCAGGCATGGTGGCTCATGCCTGTAATTTCA
GCACTTTGGGGAGGCCAAGGCAGGAGGATCACTTGAGGCCAGGAGTTGGAGACCAGCCTGGGCAACCATA
GTGAGACTCTGTCTCTACAAACATATATGTGTGTGTATATATATAAAATATATAGCGTGTGTATATATAT
ATCATATATAATATATATTGTGTATATATAATATATATATGATATAATATATACAAATGTGT
TATATATATATATAAATTAGCTGGACTTGGTGGCACATGCTCATAGTCCCAGCTACTTAGGAGACTAA
AGCAGGAGGATCACTTGAGCCCAGGAAGTTGAGGCTGAACTAAGCAATGATCCCACCTCTGCACTCCAGC
CTGGGCAGCAGAGTGACAACCTGTCTCTAGAAAAAAAAAAAAAAAAAAAAATTTAATATTATTGATTTAATATT
TTAAACATTATTTAAAAAATATTTTTAAATGTGGGAAAAAATAGAGTAACGTGAGATTTTCTCTGTGATAG
TGCTACTTAAAGCAGAATCTGAGGATAACACTGGCTGAGAACTATCACCCATCAGCAGTGAGATTAGTAC
TTAACACCTATCAGCAGCGAGATTAGTACTGAAACTGGAAGTGTTAGAAACTTATAGCAGTTCGATGTTG
CGGTGCCATCCAAGTGCGTTTTCAGCAGGCTTGTCTTATTGATCAGGTTATAGACCCATCAGGGTGTTAT
AGAACTCACATACTGAGCTCTTTGTGCTTTGTCGTGTCTCAGACATGCTCAGCAGGGCCATATGTCGG
TCCACAAGGGATTGAAAATGAAACAAACTGGTCCTTCCACCACTGATAGCTTGAGAAGAGTAGCGCTCTA
AGATGTGCTAAGTATATCTGCCCCTTTGTGGGCAAGGTACCAGAGGAGGGAGATATACGTCTGCCCCTTA
CAGCAAGGATTCCATAGCCGATGGTGTCTGGATAGAGACTGTGATAATGTTAGCCCCATTTGAAGGGGAC
GGCCACTGCTCAGCTCCAGCTGCTTGTTGCCATGTGCTGGGATATTTATGTATCCACCTAACCTTTATAT
AGCTCTTGCAATGTGTCAAACATTGTTCTGAGCACGTCATAAATATTAGCTTGCTTAATTACATTGTCAT
AACACTGTGAGGGAGGAATATTGTTATGATTCTCATTTCAGAGTTGAAGAAACAGAAATGGAGAGGTTGA
GGGACTCACCCAAAGTCACTCAGCTTTCAGAGTGGTAGAGCAGGGATTTGAACCTGTGCATATGATTTCA
GAACCTTGCTCTTAATCACACCAGGCTGCCAGTCTAATACAAGCCCCATCCTGTCAGATCTTCCAGTTTT
TCCAGAGAAGTTAAAAATGTGGATTTTTAAAAATATGAAATCTATTTCAACACTGCTAGACAAACAAAAT
GAGGCTCTGAGTTGTAGCTTGTCCATGCAGTGGGTTTTACTTTCTATCCTCCTCAAATACATCCACATCT
GTGTTCCCATTTGTCCAAGAACAAAGAGTAGATATCCTCATCCCCATGTTTCAGATGGAAAAAAAAAAAA
AAAATGAGGCCTTGGTGACTAAGCGCCTTGCCTGATGTCTTAGAAGGGAGCAATTAGTGCAGAGTGATGA
CTGCCTGCTTCCAGCCCAGGTTATGTTATTCTCGAAAGATTTATGTGCTATAATTATTTAAGAGGACAGC
AGATAAATATATACTTCAGCCTCTGAAGAAGAGTTTCTCAAAGCTAGACCACCTGCATTAGAATCATGGG
TGTGCTTGATTCAAACATAGGCTCCTGGGCCTCCCCCTAACCCCTTGCATCAGAACTCTACAGAGGTGGG
GCCCAGGAATCTGCATGTTAAGCAGATCTCTGCTGAGGCTGATGTGCACCATTGTCTGAGGGGAGATGTG
CCTGGGTTTGTCTGCTCTGACTGTATCATCCTCACGTTGTGGCTCATGAGGAAATCAGAAGGGCTAGAGG
TTGAGGAATGCTGGAAAGGGCAAGTGAGGAAGACACTCAATTTCCATTCCTCAAGGAGGGAGTGGACGCGG
TTTCCATTCCTAAAGAAGACATCATGGGAGATTTACTCTCATGATTTTCTAGGATCCTTGGGCAAAGCAA
CTAATGCCCCTTTGCCTCAGATTTTTGGGAAGCAACCCTGGCCATGCCTGATAAAACTGAGGGAAAAAAA
CTCCTGAGATCAGCACTGTCTAATATGGCAGCCATATGGGGCTGTGGAAATTTAAACGAATTAAAATTAA
ATGAAATTAAAATTCAGGCCATTAGTTGCACTAGACACATTTTAAGTACTCAACAGCAATGGCCTGAAG
TTTAAATTTATTTAATTTAATTCTTTTAAATTTCAATAGCCTCCTGTGGCTAGAGGTGACCCTGCTAG
AAGGTGCAGATGACAGAGTGAACTGATAAGATGGGCACGATATTAAGCCATCATTAGTCTCTGAAGTTCT
TACATGAGCCCTAATTTTTTGTCTTTCTAATTAATTAATAGTTAGGATTACTGGTTCTGGAGTCACACTT
GCTGGGATGAGATCAAGCCTTCATCATTTAGGAGTTGTGTGGCCTTGAACAAGTCACTTAAACTCTGCAA
AACTCAATTTCCTCATCCATGGAATTTTGTGAATAAGTGGATAAAGGTGTTCCTGTAGTACTTCCTTTGT
ATAGCTTTGGTGAGGGTTAAATGATAATTGCGTTTAAAATCATTAATATAGTCTTTGACACATATGACCT
TCTATAATGGTTACCTGCGACTTTTATTATTATTAATTCTTTCTCCTCCCAAACACACTGATTCAAGTT
TTGACCTGTTGTGGCTACTAACTTCTCCCACCATCCACCAGCTGTGCAGGTTTGCATTTTAGATTTGAAA
ATACTCCTGCATGGGCCAGGCGTGGTGGCTCACACCTGTAATCCAACACTTTGGGAGGCCAAGGCAGGT
GGATCACTTGAGGCCAGAAGTTCAAGACCAGCCTTGCCAACGTGGCAAAACCCCGTCTCTACTAAAAATA
CAGAAATTAGCCAGGCATGGTGGTGCATGACTGTAGTTCCAGCTTTTTGGGAGGCTGAGGCACAAGAATC
ACTTGAACCCAGGAGGCGGAGGTTTCAGTG

The NOV3 nucleic acid was identified on chromosome 3. This information was assigned using OMIM, the electronic northern bioinformatic tool implemented by CuraGen

Corporation, public ESTs, public literature references and/or genomic clone homologies. This was executed to derive the chromosomal mapping of the SeqCalling assemblies, Genomic clones, literature references and/or EST sequences that were included in the invention.

A disclosed NOV3 polypeptide (SEQ ID NO:11) encoded by SEQ ID NO:10 has 320 amino acid residues and is presented in Table 3B using the one-letter amino acid code. SignalP results predict that NOV3 contains no known signal peptide. Psort and/or Hydropathy results predict that NOV3 is likely to be localized extracellularly with a certainty of 0.3700. In an alternative embodiment, NOV3 is likely to be localized to the lysosome lumen with a certainty of 0.1900, or to the endoplastic reticulum membrane with a certainty of 0.1000, or to the endoplastic reticulum lumen with a certainty of 0.1000. NOV3 has a molecular weight of 35510.0 Daltons.

Table 3B. Encoded NOV3 protein sequence (SEQ ID NO:11).

```
MCFTSQCRALSSSSASPMSLIFLGIDQGQMTYDGQHWHATETCFCCAHCKKSLLGRPFLPKQGQIPCSRACSA
GEDPNGSDSSDSAFQNARAKESRRSAKIGKNKGKTEEPMLNQHSQLQVSSNRLSADVDPLSLQMDMLSLSSQT
PSLNRDPIWRSREEPYHYGNKMEQNQTQSPLQLLSQCNIRTSYSPGGQGAGAQPEMWGKHFSNPKRSSSLAMT
GHAGSFIKECREDYYPGRLRSQESYSDMSSQSFSETRGSIQVPKYEEEEEEEGGLSTQQCRTRHPISSLKYTE
DMTPTEQTPRGSMESLALSNATGRFCSP
```

The reverese complement for NOV3 is presented in Table 3C.

Table 3C. Reverse complement of the NOV3 sense strand (SEQ ID NO:12).

```
CACTGAAACCTCCGCCTCCTGGGTTCAAGTGATTCTTGTGCCTCAGCCTCCCAAAAAGCTGGAACTACAGTCATGCACCAC
CATGCCTGGCTAATTTCTGTATTTTTAGTAGAGACGGGGTTTTGCCACGTTGGCAAGGCTGGTCTTGAACTTCTGGCCTCA
AGTGATCCACCTGCCTTGGCCTCCCAAAGTGTTGAGATTACAGGTGTGAGCCACCACGCCTGGCCCATGCAGGAGTATTTT
CAAATCTAAAATGCAAACCTGCACAGCTGGTGGATGGTGGGAGAAGTTAGTAGCCACAACAGGTCAAAACTTGAATCAGTG
TGTTTGGGAGGAGAAAGAATTAATAATAATAAAAAGTCGCAGGTAACCATTATAGAAGGTCATATGTGTCAAAGACTATAT
TAATGATTTTAAACGCAATTATCATTTAACCCTCACCAAAGCTATACAAAGGAAGTACTACAGGAACACCTTTATCCACTT
ATTCACAAAATTCCATGGATGAGGAAATTGAGTTTTGCAGAGTTTAAGTGACTTGTTCAAGGCCACACAACTCCTAAATGA
TGAAGGCTTGATCTCATCCCAGCAAGTGTGACTCCAGAACCAGTAATCCTAACTATTAATTAATTAGAAAGACAAAAAATT
AGGGCTCATGTAAGAACTTCAGAGACTAATGATGGCTTAATATCGTGCCCATCTTATCAGTTCACTCTGTCATCTGCACCT
TCTAGCAGGGTCACCTCTAGCCACAGGAGGCTATTGAAATTTAAAAGAATTAAATTAAATAAAATTTAACTTCAGGCCA
TTGCTGTTGAGTACTTAAAATGTGTCTAGTGCAACTAATGGCCTGAAATTTTAATTTCATTTAATTTTAATTCGTTTAAAT
TTCCACAGCCCCATATGGCTGCCATATTAGACAGTGCTGATCTCAGGAGTTTTTTTCCCTCAGTTTTATCAGGCATGGCCA
GGGTTGCTCTCCCAAAAATCTGAGGCAAGGGCATTAGTTGCTTTGCCCAAGGATCCTAGAAAATCATGAGAGTAAATCTC
CCATGATGTCTTCTTTAGGAATGGAAACCGCGTCCACTCCCTCCTTAGGAATGGAAATTGAGTGTCTTCCTCACTTGCCCT
TTCCAGCATTCCTCAACCTCTAGCCCTTCTGATTTCCTCATGAGCCACAACGTGAGGATGATACAGTCAGAGCAGACAAAC
CCAGGCACATCTCCCCTCAGACAATGGTGCACATCAGCCTCAGCAGAATCTGCTTAACATGCAGATTCCTGGGCCCCACC
TCTGTAGAGTTCTGATGCAAGGGGTTAGGGGGAGGCCCAGGAGCCTATGTTTGAATCAAGCACACCCATGATTCTAATGCA
GGTGGTCTAGCTTTGAGAAACTCTTCTTCAGAGGCTGAAGTATATATTTATCTGCTGTCCTCTTAAATAATTATAGCACAT
AAATCTTTCGAGAATAACATAACCTGGGCTGGAAGCAGGCAGTCATCACTCTGCACTAATTGCTCCCTTCTAAGACATCAG
GCAAGGCGCTTAGTCACCAAGGCCTCATTTTTTTTTTTTTTTCCATCTGAAACATGGGGATGAGGATATCTACTCTTTGT
TCTTGGACAAATGGGAACACAGATGTGGATGTATTTGAGGAGGATAGAAAGTAAAACCCACTGCATGGACAAGCTACAACT
CAGAGCCTCATTTTGTTGTCTAGCAGTGTTGAAATAGATTTCATATTTTTAAAAATCCACATTTTAACTTCTGGGAAA
AACTGGAAGATCTGACAGGATGGGGCTTGTATTAGACTGGCAGCCTGGTGTGATTAAGAGCAAGGTTCTGAAATCATATGC
ACAGGTTCAAATCCCTGCTCTACCACTCTGAAAGCTGAGTGACTTTGGGTGAGTCCCTCAACCTCTCCATTTCTGTTTCTT
CAACTCTGAAATGAGAATCATAACAATATTCCTCCCTCACAGTGTTATGACAATGTAATTAAGCAAGCTAATAATTATGAC
GTGCTCAGAACAATGTTTGACACATTGCAAGAGCTATATAAGGTTAGGTGGATACATAAATATCCCAGCACATGGCAACA
AGCAGCTGGAGCTGAGCAGTGGCCGTCCCTTCAAATGGGGCTAACATTATCACAGTCTCTATCCAGACACCATCGGCTAT
GGAATCCTGCTGTGAAGGGCAGACGTATATCTCCCTCCTCTGGTACCTTGCCCACAAAGGGCAGATATACTTAGCACAT
CTTAGAGCGCTACTCTTCTCAAGCTATCAGTGGTGAAGGACCAGTTTGTTTTCATTTTCAATCCCTTGTGGACCGACATAT
GGCCCTGCTGAGCATGTCTGAGACACAGCACAAAGCACAAAGAGCTCAGTATGTGAGTTCTATAACACCCTGATGGGTCTA
TAACCTGATCAATAAGACAAGCCTGCTGAAAACGCACTTGGATGGCACCGCAACATCGAACTGCTATAAGTTTCTAACACT
TCCAGTTTCAGTACTAATCTCGCTGCTGATAGGTGTTAAGTACTAATCTCACTGCTGATGGGTGATAGTTCTCAGCCAGTG
TTATCCTCAGATTCTGCTTTAAGTAGCACTATCACAGAGAAAATCTACGTTACTCTATTTTTTCCCACATTTAAAAATATT
```

The full NOV3 amino acid sequence of the protein of the invention was found to have 59 to 120 amino acid residues (49%) identical to, and 80 to 120 amino acid residues (66%) similar to, the 1011 amino acid residue SPTREMBL-ACC:Q9NDQ8 PRICKLE 2 from Ciona intestinalis. In additional searches of the public databases, NOV3 has homology to the amino acid sequences shown in the BLASTP data listed in Table 3D.

Table 3D. BLAST results for NOV3

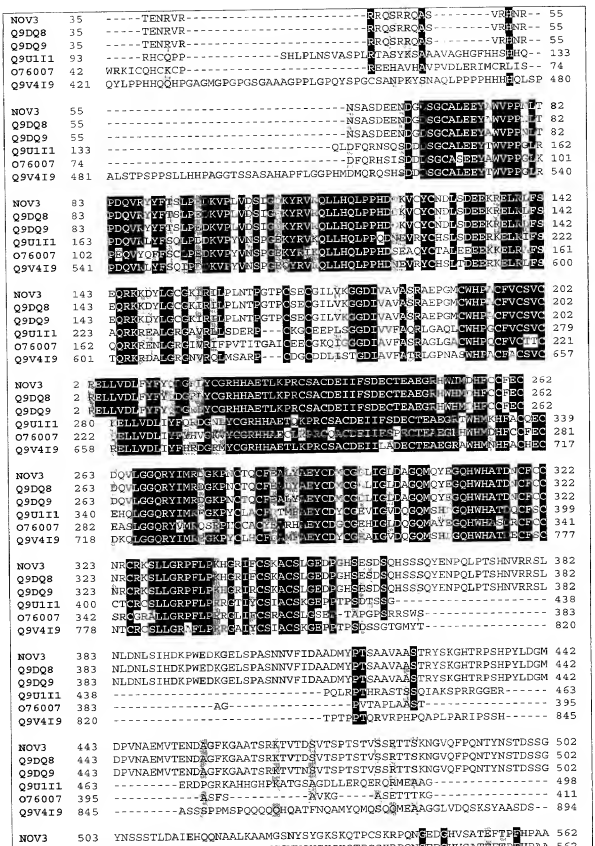
Matching Entry (in SwissProt + SpTrEMBL)	Description	Length (aa)	Identity (%)	Positives (%)	Expect
Q9NDQ8; AB036841; BAB00618.1	PRICKLE 2. ciona intestinalis 6/2001	1011	59/122 (48%)	78/122 (64%)	1e-23
Q9NDQ9; AB036840; BAB00617.1	PRICKLE 1. ciona intestinalis. prickle 1 6/2001	1066	58/122 (48%)	77/122 (63%)	1e-22
Q9U1I1; AJ251892; CAB64381.1	LIM-DOMAIN PROTEIN (ESN PROTEIN). drosophila melanogaster 6/2001	785	47/89 (53%)	60/89 (67%)	2e-20
O76007; AJ011654; CAA09726.1	TRIPLE LIM DOMAIN PROTEIN. homo sapiens 6/2001	615	38/61 (62%)	49/61 (80%)	4e-20
Q9V4I9; AE003842; AAF59281.1	CG11084 PROTEIN drosophila melanogaster 6/2001	1268	47/105 (45%)	62/105 (59%)	8e-20

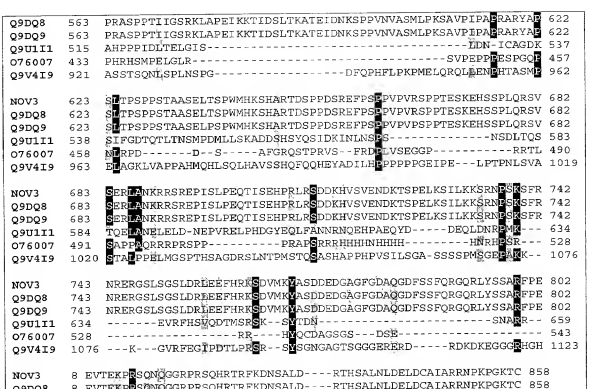
The homology of these and other sequences is shown graphically in the ClustalW analysis shown in Table 3E. In the ClustalW alignment of the NOV3 protein, as well as all other ClustalW analyses herein, the black outlined amino acid residues indicate regions of conserved sequence (*i.e.*, regions that may be required to preserve structural or functional properties), whereas non-highlighted amino acid residues are less conserved and can potentially be mutated to a much broader extent without altering protein structure or function.

Table 3E. ClustalW Analysis of NOV3

1) Novel NOV3 (SEQ ID NO:11)
2) Q9DQ8 (SEQ ID NO:13)
3) Q9DQ9 (SEQ ID NO:14)
4) Q9U1I1 (SEQ ID NO:15)
5) O76007 (SEQ ID NO:16)
6) Q9V4I9 c-ter fragment (SEQ ID NO:17)

```
NOV3          1   ------------------------------------------------------MTMPAAATEQ 10
Q9DQ8         1   ------------------------------------------------------MTMPAAATEQ 10
Q9DQ9         1   ------------------------------------------------------MTMPAAATEQ 10
Q9U1I1        1   -------MQQAP-------QQQQHPHP----PSSSYYTQTES--------ELLQIPAGGTGL 36
O76007        1   ------------------------------------------------------MFARGSRRRR 10
Q9V4I9      241   EEESPEQEAPKPALPPKQKQQRPVPPLPPPPANRVTQDQGTQPAAPQVPLQPETAGDLQF 30

NOV3         11   TRGTMPSNIDPAG-------------AGLDQDIVIRGA---------------------- 35
Q9DQ8        11   TRGTMPSNIDPAG-------------AGLDQDIVIRGA---------------------- 35
Q9DQ9        11   TRGTMPSNIDPAG-------------AGLDQDIVIRGA---------------------- 35
Q9U1I1       37   TFASHSQR--PAG-------------AISQVASTAHLDVAAASS---------------- 66
O76007       11   SGRAPPEAEDADR-------------GQPCNSCREQQAGFLLHG---------------- 41
Q9V4I9      301   LNLSLRQRSLPAGMKPFKDAHDISFTPNELDTSAEPEVATGAAQQESNECRTPLTQISYL 360

NOV3         35   ------------------------------------------------------------ 35
Q9DQ8        35   ------------------------------------------------------------ 35
Q9DQ9        35   ------------------------------------------------------------ 35
Q9U1I1       66   ----------------------GSGGSAVSGGSGG-----APESAGRFVS---PLQR-- 93
O76007       41   ------------------------------------------------------------ 41
Q9V4I9      361   QKIPTLPRHFSPSGQGLATPPALGSGGMGLPSSSSASALYAAQAAAGILPTSPLPLQRHQ 420
```





Other BLAST results include sequences from the Patp database, which is a proprietary database that contains sequences published in patents and patent publications. Patp results include those listed in Table 3F.

Table 3F. Patp Alignment of NOV3

Sequences producing High-scoring Segment Pairs	Protein/ Organism	Length (aa)	Identity (%)	Positive (%)	Expect
patp:AAW83952	Polypeptide encoded by gene 2 clone HDTAY29-*H. sapiens*	159	44	67	1.4e-07
Patp:AAY57563	Human testin (HTES)-H. sapiens	421	44	67	3.4e-05
patp.AAB93751	Human protein SEQ ID NO:13416-*H. sapiens*	464	44	67	3.4e-05
Patp:AAB42119	Human ORFX ORF1883 polypeptide-H. Sapiens	464	44	67	4.0e-05
Patp:AAG01529	Human secreted protein-H. sapiens	126	30	44	5.8e-05
Patp AAY84378	Amino acid sequence of a human LIM domain protein homologue-H. sapiens	280	32	50	0.00077

The results of a domain search indicate that the NOV3 protein contains the protein domain (as defined by Interpro) named IPR001781 at amino acid positions 43 to 76. Table 3G lists the domain description from further DOMAIN analysis results against NOV3. This indicates that NOV3 has properties similar to those of other proteins known to contain these domains and similar to the properties of these domains.

Table 3G. Domain Analysis of NOV3

```
PRODOM ANALYSIS
Sequences producing High-scoring Segment Pairs:                    Smallest
                                                          High     Sum
                                                          Score    Probability
                                                                   P(N)

Nprdm:21599 p36 (1) TES2_MOUSE // TESTIN 2 (TES2) (CONTAIN ..     127  1.8e-08
prdm:39635 p36 (1) ZYX_MOUSE // ZYXIN. REPEAT; LIM MOTIF;. .      68  0.048
prdm:67    p36 (155) LIM1(10) LIM3(8) PAXI(8)  // PROTEIN. . .    67  0.061
prdm:55854 p36 (1) HMW1_MYCGE // CYTADHERENCE HIGH MOLECU. .      72  0.15
prdm:7588  p36 (3) SLI3(2) LRG1(1)  // PROTEIN LIM MOTIF ...      61  0.25

prdm:21599 p36 (1) TES2_MOUSE // TESTIN 2 (TES2) (CONTAINS  TESTIN 1 (TES1)). LIM
MOTIF; METAL-BINDING; ZINC; ALTERNATIVE SPLICING, 66 aa.
Expect = 1.8e-08, Identities = 19/43 (44%), Positives = 29/43 (67%)
for NOV3 aa residues 29 to 71; and LIM Domain residues 19 to 61

>prdm:39635 p36 (1) ZYX_MOUSE // ZYXIN. REPEAT; LIM MOTIF; METAL-BINDING; ZINC;
CELL ADHESION, 44 aa.
 Identities = 13/34 (38%), Positives = 19/34 (55%)

>prdm:67 p36 (155) LIM1(10) LIM3(8) PAXI(8) // PROTEIN LIM MOTIF METAL-BINDING
ZINC REPEAT HOMEOBOX NUCLEAR DNA-BINDING DEVELOPMENTAL, 68 aa.
 Identities = 14/37 (37%), Positives = 20/37 (54%)

>prdm:55854 p36 (1) HMW1_MYCGE // CYTADHERENCE HIGH MOLECULAR WEIGHT PROTEIN 1
(CYTADHERENCE ACCESSORY PROTEIN 1). STRUCTURAL PROTEIN, 107 aa.
 Identities = 18/67 (26%), Positives = 37/67 (55%)

>prdm:7588 p36 (3) SLI3(2) LRG1(1) // PROTEIN LIM MOTIF METAL-BINDING ZINC REPEAT
SKELETAL MUSCLE LIM-PROTEIN SLIM, 67 aa.
 Identities = 20/55 (36%), Positives = 30/55 (54%)

BLOCKS ANALYSIS
AC#       Description                                  Strength  Score  AA#
BL00115R Eukaryotic RNA polymerase II heptapeptide rep  2074   1110  124
```

```
BL00911C Dihydroorotate dehydrogenase proteins.          1314   1050  201
BL01137D Uncharacterized protein family UPF0006 protei  1297   1048  126
BL00576B General diffusion Gram-negative porins protei  1391   1047  172
BL01182C Glycosyl hydrolases family 35  proteins.       1577   1046   73

ProSite Analysis                                              NOV3 aa position
Pattern-ID: ASN_GLYCOSYLATION PS00001 (Interpro)                  78, 171, 312
Pattern-DE: N-glycosylation site, Pattern: N[^P][ST][^P]

Pattern-ID: CAMP_PHOSPHO_SITE PS00004 (Interpro)                          211
Pattern-DE: cAMP- and cGMP-dependent protein kinase phosphorylation site
Pattern:    [RK]{2}.[ST]

Pattern-ID: PKC_PHOSPHO_SITE PS00005 (Interpro)      95, 98, 123, 287, 300, 314
Pattern-DE: Protein kinase C phosphorylation site
Pattern:    [ST].[RK]

Pattern-ID: CK2_PHOSPHO_SITE PS00006 (Interpro)         72, 157, 243, 251, 295
Pattern-DE: Casein kinase II phosphorylation site
Pattern:    [ST].{2}[DE]

Pattern-ID: TYR_PHOSPHO_SITE PS00007 (Interpro)                   156, 227
Pattern-DE: Tyrosine kinase phosphorylation site
Pattern:    [RK].{2,3}[DE].{2,3}Y

Pattern-ID: MYRISTYL PS00008 (Interpro)              24, 63, 79, 192, 272,  303
Pattern-DE: N-myristoylation site
Pattern:    G[^EDRKHPFYW].{2}[STAGCN][^P]

Pattern-ID: LEUCINE_ZIPPER PS00029 (Interpro)                             119
Pattern-DE: Leucine zipper pattern
Pattern:    L.{6}L.{6}L.{6}L
```

The LIM domain is a zinc finger structure that is present in several types of proteins, including homeodomain transcription factors, kinases and proteins that consist of several LIM domains. Proteins containing LIM domains have been discovered to play important roles in a variety of fundamental biological processes including cytoskeleton organization, cell lineage specification and organ development, but also for pathological functions such as oncogenesis, leading to human disease. The LIM domain has been demonstrated to be a protein-protein interaction motif that is critically involved in these processes. The recent isolation and analysis of more LIM domain-containing proteins from several species have confirmed and broadened our knowledge about LIM protein function. Furthermore, the identification and characterization of factors that interact with LIM domains illuminates mechanisms of combinatorial developmental regulation.

LIM domain containing proteins generally have two tandem copies of a domain, called LIM (for Lin-11 Isl-1 Mec-3) in their N-terminal section. Zyxin and paxillin are exceptions in that they contains respectively three and four LIM domains at their C-terminal extremity. In apterous, isl-1, LH-2, lin-11, lim-1 to lim-3, lmx-1 and ceh-14 and mec-3 there is a homeobox domain some 50 to 95 amino acids after the LIM domains. In the LIM domain, there are seven conserved cysteine residues and a histidine. The arrangement followed by these conserved residues is C-x(2)-C-x(16,23)-H-x(2)-[CH]-x(2)-C-x(2)-C-x(16,21)-C-x(2,3)-[CHD].

The LIM domain binds two zinc ions. LIM does not bind DNA, rather it seems to act as interface for protein-protein interaction.

The *Prickle* gene in *Drosophila* belongs to a family of "tissue polarity" genes that control the orientation of bristles and hairs in the adult cuticle. (*See* Gubb and Garcia-Bellido, J. *Embryol. Exp. Morphol.* 68:37-57 (1982)) These "tissue polarity" genes play important roles in the organization of the cytoskeleton. Prickle has been shown to be involved in hereditary benign intraepithelial dyskeratosis (OMIM Entry: 127600). Characteristic histologic changes of the prickle cell layer of the mucosa include numerous round, waxy-looking, eosinophilic cells that appear to be engulfed by normal cells. The conjunctiva and oral mucous membranes are affected. The oral lesion, which grossly resembles leukoplakia, is not precancerous. The eye lesions resemble pterygia (see OMIM 178000). The only symptoms are produced by involvement of the cornea, resulting in impairment of vision.

The human homolog of Drosophila discs large-3 (DLG3) is a protein related to Prickle and LIM. See, OMIM Entry 300189. Mutations of the 'discs large' (dlg) tumor suppressor locus in Drosophila lead to imaginal disc neoplasia and a prolonged larval period followed by death. Drosophila dlg and related proteins form a subfamily of the membrane-associated guanylate kinase (MAGUK) protein family and are important components of specialized cell junctions. See DLG1 (OMIM 601014). A partial cDNA encoding NEDLG (neuroendocrine DLG) was isolated by searching an EST database for sequences related to dlg and DLG1. See, Makino *et al.* (1997). Northern blot analysis revealed that NEDLG is highly expressed in neuronal and endocrine tissues. Immunolocalization studies indicated that the protein was expressed mainly in nonproliferating cells, such as neurons, cells in Langerhans islets of the pancreas, myocytes of heart muscles, and the prickle and functional layer cells of the esophageal epithelium. In a yeast 2-hybrid assay, NEDLG interacted with the C-terminal region of the APC (OMIM 175100) tumor suppressor protein. Therefore, NEDLG may negatively regulate cell proliferation through its interaction with the APC protein. By fluorescence in situ hybridization, Makino *et al.* (1997) mapped the NEDLG gene to Xq13. Using radiation hybrid panels, Stathakis *et al.* (1998) refined the map position to Xq13.1. DLG3 is located within the dystonia-parkinsonism syndrome (DYT3; OMIM 314250) locus.

The disclosed NOV3 nucleic acid encoding a LIM-domain-containing Prickle-like secreted protein includes the nucleic acid whose sequence is provided in Table 3A, or a fragment thereof. The invention also includes a mutant or variant nucleic acid any of whose bases may be changed from the corresponding base shown in Table 3A while still encoding a protein that maintains its LIM-domain-containing Prickle-like activities and physiological functions, or a fragment of such a nucleic acid. The invention further includes nucleic acids whose sequences

are complementary to those just described, including nucleic acid fragments that are complementary to any of the nucleic acids just described. The invention additionally includes nucleic acids or nucleic acid fragments, or complements thereto, whose structures include chemical modifications. Such modifications include, by way of nonlimiting example, modified bases, and nucleic acids whose sugar phosphate backbones are modified or derivatized. These modifications are carried out at least in part to enhance the chemical stability of the modified nucleic acid, such that they may be used, for example, as antisense binding nucleic acids in therapeutic applications in a subject. In the mutant or variant nucleic acids, and their complements, up to about 17 % percent of the bases may be so changed.

The disclosed NOV3 protein of the invention includes the LIM-domain-containing Prickle-like protein whose sequence is provided in Table 3B. The invention also includes a mutant or variant protein any of whose residues may be changed from the corresponding residue shown in Table 3B while still encoding a protein that maintains its LIM-domain-containing Prickle-like activities and physiological functions, or a functional fragment thereof. In the mutant or variant protein, up to about 16 % percent of the residues may be so changed.

The invention further encompasses antibodies and antibody fragments, such as Fab, (Fab)2 or single chain FV constructs, that bind immunospecifically to any of the proteins of the invention. Also encompassed within the invention are peptides and polypeptides comprising sequences having high binding affinity for any of the proteins of the invention, including such peptides and polypeptides that are fused to any carrier partcle (or biologically expressed on the surface of a carrier) such as a bacteriophage particle.

The protein similarity information, expression pattern, and map location for the novel LIM-domain-containing Prickle-like NOV3 protein and nucleic acid disclosed herein suggest that this novel LIM-domain-containing Prickle-like protein may have important structural and/or physiological functions characteristic of the LIM-domain-containing Prickle-like protein family. For example, NOV3 may be important for the proper organization of cytoskeleton, or in the treatment of dystonia-parkinsonism syndrome; hereditary benign intraepithelial dyskeratosis; developmental disorders and other diseases, disorders and conditions of the like. Accordingly, NOV3 nucleic acids and proteins may have potential diagnostic and therapeutic applications in treating disorders that involve cytoskeleton malfunctions. These include serving as a specific or selective nucleic acid or protein diagnostic and/or prognostic marker, wherein the presence or amount of the nucleic acid or the protein are to be assessed, as well as potential therapeutic applications such as the following: (i) a protein therapeutic, (ii) a small molecule drug target, (iii) an antibody target (therapeutic, diagnostic, drug targeting/cytotoxic antibody), (iv) a nucleic acid

useful in gene therapy (gene delivery/gene ablation), and (v) a composition promoting tissue regeneration in vitro and in vivo (vi) biological defense weapon.

Based on the tissues in which NOV3 is most highly expressed, including kidney and ovary, specific uses include developing products for the diagnosis or treatment of a variety of diseases and disorders. Additional disease indications and tissue expression for NOV3 is presented in Example 2.

The nucleic acids and proteins of the invention are useful in potential diagnostic and therapeutic applications implicated in, but not limited to, various diseases and disorders described below and/or other pathologies. For example, the compositions of the present invention will have efficacy for treatment of patients suffering from: dystonia-parkinsonism syndrome; dyskeratosis, hereditary benigh intraepithelial; developmental disorders and other diseases, disorders and conditions of the like. A cDNA encoding the LIM-domain-containing Prickle-like protein NOV3 may be useful in gene therapy, and the Prickle-like protein NOV3 may be useful when administered to a subject in need thereof.

These materials are further useful in the generation of antibodies that bind immunospecifically to the novel substances of the invention for use in therapeutic or diagnostic methods.

NOV3 nucleic acids and polypeptides are further useful in the generation of antibodies that bind immuno-specifically to the novel NOV3 substances for use in therapeutic or diagnostic methods. These antibodies may be generated according to methods known in the art, using prediction from hydrophobicity charts, as described in the "Anti-NOVX Antibodies" section below. The disclosed NOV3 protein has multiple hydrophilic regions, each of which can be used as an immunogen. In one embodiment, a contemplated NOV3 epitope is from about amino acids 25 to 50. In another embodiment, a NOV3 epitope is from about amino acids 55 to 140. In additional embodiments, NOV3 epitopes are from about amino acids 145 to 180, from about amino acids 180 to 225, and from about amino acids 250 to 280. These novel proteins can be used in assay systems for functional analysis of various human disorders, which will help in understanding of pathology of the disease and development of new drug targets for various disorders.

NOV4

A disclosed NOV4 nucleic acid of 1278 nucleotides (also referred to as CG56824-01) encoding a novel lipid metabolism-like protein is shown in Table 4A. An open reading frame was identified beginning with an ATG initiation codon at nucleotides 184 to 186 and ending with a TGA codon at nucleotides 1195 to 1197. Putative untranslated regions upstream from the

initiation codon and downstream from the termination codon are underlined in Table 4A, and the start and stop codons are in bold letters.

Table 4A. NOV4 nucleotide sequence (SEQ ID NO:18).

CTCTTCGTGGCCCAACGCCCCCAATCCTTGCGTGTCCTTGCAGTCCCACCCCACACTCAGCCTTGTGTCCCTCGATCCAGT
CTCCGACTTCCATTTCCCCACCCTAAACCGCCTACCCGGGTGTCTGTTCCCCGCCCGGTTGTCCTCGCCCTGCTGCGCTGAG
TGTCCCCTGTTAGCCTCGACCCC**ATG**GCGCTGCAGACGCTGCAGAGCTCGTGGGTGACCTTCCGCAAGATCCTGTCTCAC
TTCCCCGAGGAGCTGAGTCTGGCTTTCGTCTACGGCTCCGGGGTGTACCGCCAGGCAGGGCCCAGTTCAGACCAGAAGAA
TGCTATGCTGGACTTTGTGTTCACAGTAGATGACCCTGTCGCATGGCATTCAAAGAACCTGAAGAAAAATTGGAGTCACT
ACTCTTTCCTAAAAGTTTTAGGGCCCAAGATTATCACGTCCATCCAGAATAACTATGGCGCTGGAGTTTACTACAATTCA
TTGATCATGTGTAATGGTAGGCTTATCAAATATGGAGTTATTAGCACTAACGTTCTGATTGAAGATCTCCTCAACTGGAA
TAACTTATACATTGCTGGACGACTCCAAAAACCGGTGAAAATTATCTCAGTGAACGAGGATGTCACTCTTAGATCAGCCC
TCGATAGAAATCTGAAGAGTGCTGTGACCGCTGCTTCCTCATGCTCCCCGAAAGCTTTTCTGAAGAAGACCTCTTCATA
GAGATTGCCGGTCTCTCCTATTCAGGTGACTTTCGGATGGTGGTTGGAGAAGATAAAACAAAAGTGTTGAATATTGTGAA
GCCCAATATAGCCCACTTTCGAGAGCTCTATGGCAGCATACTACAGGAAAATCCTCAAGTGGTGTATAAAAGCCAGCAAG
GCTGGCTGGAGATAGATAAAAGCCCAGAAGGACAGTTCACTCAGCTGATGACATTGCCCAAAACCTTACAGCAACAGATA
AATCATATTATGGACCCTCCTGGAAAAAACAGAGATGTGGAAGAAACTTTATTCCAAGTGGCTCATGATCCCGACTGTGG
AGATGTGGTGCGACTAGGGCTTTCAGCAATCGTGAGACCGTCTAGTATAAGACAGAGCACGAAAGGCATTTTTACTGCTG
GCCTGAAGAAGTCAGTGATTTATAGTTCACTAAAACTGCACAAAATGTGGAAAGGGTGGCTGAGGAAACATCC**TGA**TTT
TGCTTGCTTTTATATATGTTATGTGTAGATGAATAAAGTGTTTGATCCTTTTTGACAAAAAAAAAAAAAAAAAAAAAA

In a search of public sequence databases, the NOV4 nucleic acid sequence has 96 of 101 bases (95 %) identical to a human cDNA clone NT2RP3003346. Public nucleotide databases include all GenBank databases and the GeneSeq patent database.

A disclosed NOV4 polypeptide (SEQ ID NO:19) encoded by SEQ ID NO:18 has 337 amino acid residues and is presented in Table 4B using the one-letter amino acid code. SignalP, Psort and/or Hydropathy results predict that NOV4 has a signal peptide. The most likely cleavage site is between amino acid positions 14 and 15, *i.e.*, at the dash between TFR-KI. NOV4 is likely to be localized to the mitochondrial matrix space with a certainty of 0.6567. In alternative embodiments, NOV4 is localized to the mitochondrial inner membrane with a certainty of 0.3497, to the mitochondrial intermembrane space with a certainty of 0.3497, or the mitochondrial outer membrane with a certainty of 0.3497. NOV4 has a molecular weight of 38,078.6 Daltons.

Table 4B. Encoded NOV4 protein sequence (SEQ ID NO:19).

MALQTLQSSWVTFRKILSHFPEELSLAFVYGSGVYRQAGPSSDQKNAMLDFVFTVDDPVAWHSKNLKK
NWSHYSFLKVLGPKIITSIQNNYGAGVYYNSLIMCNGRLIKYGVISTNVLIEDLLNWNNLVIAGRLQK
PVKIISVNEDVTLRSALDRNLKSAVTAAFLMLPESFSEEDLFIEIAGLSYSGDFRMVVGEDKTKVLNI
VKPNIAHFRELYGSILQENPQVVYKSQQGWLEIDKSPEGQPTQLMTLPKTLQQQINHIMDPPGKNRDV
EETLFQVAHDPDCGDVVRLGLSAIVRPSSIRQSTKGIFTAGLKKSVIYSSLKLHKMWKGWLRKTS

The NOV4 nucleic acid was tentatively localized to human chromosome 3. The cDNA coding for the NOV4 sequence was cloned by the polymerase chain reaction (PCR) using the primer set NOV4-2, shown in Table 17A. The PCR product derived by exon linking, covering

the entire NOV4 open reading frame, was cloned into the pCR2.1 vector from Invitrogen to provide clone 110189::COR24SC128.698230.M23.

The reverse complement for NOV4 is presented in Table 4C.

Table 4C. NOV4 reverse complement (SEQ ID NO:20)

```
TTTTTTTTTTTTTTTTTTTTGTCAAAAAGGATCAAACACTTTATTCATCTACACATAACATATATAAAAGCAAGCA
AAATCAGGATGTTTTCCTCAGCCACCCTTTCCACAATTTTGTTCAGTTTTAGTGAACTATAAATCACTGACTTCTTCAG
GCCAGCAGTAAAAATGCCTTTCGTGCTCTGTCTTATACTAGACGGTCTCACGATTGCTGAAAGCCCTAGTCGCACCAC
ATCTCCACAGTCGGGATCATGAGCCACTTGGAATAAAGTTTCTTCCACATCTCTGTTTTTTCCAGGAGGGTCCATAAT
ATGATTTATCTGTTGCTGTAAGGTTTTGGGCAATGTCATCAGCTGAGTGAACTGTCCTTCTGGGCTTTTATCTATCTC
CAGCCAGCCTTGCTGGCTTTTATACACCACTTGAGGATTTTCCTGTAGTATGCTGCCATAGAGCTCTCGAAAGTGGGC
TATATTGGGCTTCACAATATTCAACACTTTTGTTTTATCTTCTCCAACCACCATCCGAAAGTCACCTGAATAGGAGAG
ACCGGCAATCTCTATGAAGAGGTCTTCTTCAGAAAAGCTTTCGGGGAGCATGAGGAAAGCAGCGGTCACAGCACTCTT
CAGATTTCTATCGAGGGCTGATCTAAGAGTGACATCCTCGTTCACTGAGATAATTTTCACCGGTTTTTGGAGTCGTCC
AGCAATGTATAAGTTATTCCAGTTGAGGAGATCTTCAATCAGAACGTTAGTGCTAATAACTCCATATTTGATAAGCCT
ACCATTACACATGATCAATGAATTGTAGTAAACTCCAGCGCCATAGTTATTCTGGATGGACGTGATAATCTTGGGCCC
TAAAACTTTTAGGAAAGAGTAGTGACTCCAATTTTTCTTCAGGTTCTTTGAATGCCATGCGACAGGGTCATCTACTGT
GAACACAAAGTCCAGCATAGCATTCTTCTGGTCTGAACTGGGCCCTGCCTGGCGGTACACCCCGGAGCCGTAGACGAA
AGCCAGACTCAGCTCCTCGGGGAAGTGAGACAGGATCTTGCGGAAGGTCACCCACGAGCTCTGCAGCGTCTGCAGCGC
CATGGGGTCGAGGCTAACAGGGGACACTCAGCGCAGCAGGGCGAGGACAACCGGGCGGGGAACAGACACCGGGTAGGC
GGTTTAGGGTGGGAAATGGAAGTCGGAGACTGGATCGAGGGACACAAGGCTGAGTGTGGGGTGGGACTGCAAGGACAC
GCAAGGATTGGGGCGTTGGGCCACGAAGAG
```

In a search of public sequence databases, the NOV4 amino acid sequence has 90 of 214 amino acid residues (42%) identical to, and 137 residues (214%) positive with, the 274 amino acid residue *C. elegans* Y71F9B.2 protein. Public amino acid databases include the GenBank databases, SwissProt, PDB and PIR.

It was also found that NOV4 had homology to the amino acid sequences shown in the BLASTP data listed in Table 4D.

Table 4D. BLAST results for NOV4

Gene Index/ Identifier	Protein/ Organism	Length (aa)	Identity (%)	Positives (%)	Expect
Q9CW36; AK005100; BAB23818.1	1500001M20RIK PROTEIN (FRAGMENT). mus musculus. 6/2001	367	271/332 (82%)	304/332, (92%)	1e-160
O74339; AL031174; CAA20110.1	HYPOTHETICAL 44.3 KDA PROTEIN C1A4.06C IN CHROMOSOME II. schizosaccharomyces pombe. 3/2001	383	119/325 (37%)	174/325, (54%)	2e-47
Q9N4G7; AC024201; AAF36018.1	Y71F9B.2 PROTEIN caenorhabditis elegans. 10/2000	274	111/320 (35%)	169/320, (53%)	5e-47
Q9VFF2; AE003706; AAF55108.1	CG3641 PROTEIN. drosophila melanogaster. 5/2000	647	109/269 (41%)	152/269, (57%)	2e-44
Q9SN75; AL132955; CAB61989.1	HYPOTHETICAL 37.4 KDA PROTEIN. arabidopsis thaliana. 5/2000	332	102/314 (32%)	170/314, (54%)	7e-41

The homology of these and other sequences is shown graphically in the ClustalW analysis shown in Table 4E. In the ClustalW alignment of the NOV4 protein, as well as all other

ClustalW analyses herein. the black outlined amino acid residues indicate regions of conserved sequence (*i.e.*, regions that may be required to preserve structural or functional properties), whereas non-highlighted amino acid residues are less conserved and can potentially be mutated to a much broader extent without altering protein structure or function.

Table 4E. ClustalW Analysis of NOV4

```
1)  NOV4 (SEQ ID NO:19)
2)  Q9CW36 (SEQ ID NO:21)
3)  O74339 (SEQ ID NO:22)
4)  Q9N4G7 (SEQ ID NO:23)
5)  Q9VFP2 (SEQ ID NO:24)
6)  Q9SN75 (SEQ ID NO:25)

NOV4      1  ------------------GTGRKRGPHDRELRAQGRHSTVCPTGGPPAHGAAGLHSSGVG 42
Q9CW36    1  ------------------GTGRKRGPHDRELRAQGRHSTVCPTGGPPAHGAAGLHSSGVG 42
O74339    1  MIFGKTHPLSYNILRYSTKRWMNRHSYSHHAKCTVAQLLKQNLLTFENQRIQPEEELKEN 60
Q9N4G7    1  ------------------------------------------------------MDEY 4
Q9VFP2    1  ------------------------------------------------------MLDLY 5
Q9SN75    1  ------------------------------------------------------METTQKD 7

NOV4     43  LRRILAHPFEDLSLAFAYGSAVYRQAGPSAHQEN--------PLLLVFTVDDPVAWTAM 94
Q9CW36   43  LRRILAHPFEDLSLAFAYGSAVYRQAGPSAHQEN--------PLLLVFTVDDPVAWTAM 94
O74339   61  LTKVVNYFQAPIDVAVGYGSGVFRQAGYSQKEN--------PVIPFIFCVDDPVKWIKI 111
Q9N4G7    5  RELISVLPLETVEYAFAYGSGAIQQQNEDKSEK---------QVEFVIVTKNAQEFERD 54
Q9VFP2    6  RRTVARFPLGSVSYMFAYGSGVKQQEGYGKVGNGNNLRPPPGTVVELVFCVDDARGEAE 65
Q9SN75    8  ELSSFLSVLPPVDFCCVYGSTLHPNNQ-DKSR----------QVEYYLGVSDPIKWESA 55

NOV4     95  NLKKNWSHYSS--PLKLIGRIISSTQNNYQAVVRNPLIRCDGK--LHKKGVDLGTGTLIE 150
Q9CW36   95  NLKKNWSHYSS--PLKLIGRIISSTQNNYQAVVRNPLIRCDGK--LHKKGVDLGTGTLIE 150
O74339  112  NLQQNPSHYSS--PVKNPGEGFVSTLQESPGQGVVQNTHVEVEGN--IHKYGVDLGKDVYE 167
Q9N4G7   55  NILKNQPGHYS--LLRLMGEKMIEKIQCNFAARVVYNTHVKVGKR--KKKYGVESYENVKQ 110
Q9VFP2   66  NLHRHPDHYS--ALRHIGENFVAKVQERLQAGVVQNTLVPLPDVGITLKKYGVVSQEELLE 123
Q9SN75   56  NIKKWSDHYASWMVHLGSARLETNVADKVQVGVHPNPFMWNNDR--KKKYGVVRMHDLVQ 113

NOV4    151  DLLAWNNLVIAGRLQKEVK-IVSMNENMALRAALDKNQRSAVTTAGLMLPSGSSEEDLFI 209
Q9CW36  151  DLLAWNNLVIAGRLQKPVK-IVSMNENMALRAALDKNQRSAVTTAGLMLPSGSSEEDLFI 209
O74339  168  DLKNWNTMVLAGRFQKPVV-ILKGEDE--FYKENSYNLSSATHVGLLMLPGRTEFDLYK 224
Q9N4G7  111  DLLDWRWIYISGRLQKPVLEVIKPRQD--MCDLVTENNRSANHHSILDLPPSSTLKQLFH 168
Q9VFP2  124  DLLDWRHLVLAGKLQKPVTNLVNPSDNPPIKAALERNLVSAHQVALLDLPGKSTAYGLFH 183
Q9SN75  114  DLLDWKRFVLSGRLQKPVHMLVDNLD---IEDVNSVNKRAAESAALDLPSKRTEEDLYA 170

NOV4    210  EHAGLSYSGLDRM-VIGGEKSKYLNNIVKPNVGHGRELMESIIQKDPQVVYKMHQG----- 263
Q9CW36  210  EHAGLSYSGLDRM-VIGGEKSKYLNNIVKPNVGHGRELMESIIQKDPQVVYKMHQG----- 263
O74339  225  TIVSLSYSGLDRMSFFAGNPKKYENIVSKQIAPGRKLMLPLFYAEPG-VHPIESSE---- 279
Q9N4G7  169  KIVGLSYTGLDRM-VVGGDKNKINKIVEGNYEELLRVMEP---------------------- 207
Q9VFP2  184  TLAGLSYKGLDRM-IPGGNKQKYHNIVSPQINDFPALMQPSLGQLSDYVAVNMKGQEPGS 242
Q9SN75  171  KICSLSYMGLDRM-FFADDTNKVNKIVKGQFDLPQSMVKPPLEECETKNLLRFSSAEAS- 228

NOV4    263  -----QLEIDKSPEGQFTQLMTLPRTLQCQIN---------------------------- 290
Q9CW36  263  -----QLEIDKSPEGQFTQLMTLPRTLQCQIN---------------------------- 290
O74339  279  ----VLKSMDPSDNSRYLSFHQN--ITKDSIS---------------------------- 305
Q9N4G7  207  -----LMNDDC-------------------ARLS---------------------------- 216
Q9VFP2  243  RKPAIIFEQCKSSSATCQHLRQLPRELQKRLQRNAACRGDYTQVVNHLSMASQLPEVLQA 302
Q9SN75  228  ---HTKLVQLSSLSATRSLVSSLPASVRSQMG-------------------------KS 259

NOV4    290  ----HLMD-------------------------------------------PPGRNRD---- 301
Q9CW36  290  ----HLMD-------------------------------------------PPGRNRD---- 301
O74339  305  ----RLLN-------------------------------------------GLPLN---- 314
Q9N4G7  216  ----VIPS-------------------------------------------LAPRH---- 225
Q9VFP2  303  SVNDIIMSSDDNSSDSNSSSDERQRKRKLKKHSKDVDKSKKKKSKKHKKEKPRHKEKKRS 362
Q9SN75  260  LGEKKFVS-------------------------------------------ETGRVMG---- 274

NOV4    301  ----------VEETLLQVAQDPDCGDVVRLAIS---------------------------S 325
Q9CW36  301  ----------VEETLLQVAQDPDCGDVVRLAIS---------------------------S 325
O74339  314  ----------LVKILGLKPDTSSFEKCAELMLTN---------------------------Q 339
```

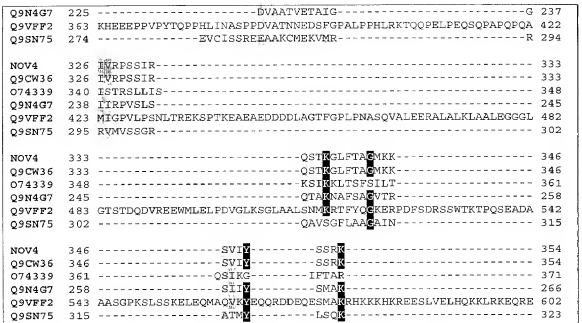


Table 4F lists the domain description from DOMAIN analysis results against NOV4. This indicates that the NOV4 sequence has properties similar to those of other proteins known to contain this domain.

Table 4F. Domain Analysis of NOV4

```
ProDom Protein Domain Analysis
prdm:50749 p36 (1) YG1W_YEAST // HYPOTHETICAL 44.2 KD PROTEIN IN RME1-TFC4
INTERGENIC REGION. HYPOTHETICAL PROTEIN, 385 aa.
Expect = 2.1e-41, Identities = 85/209 (40%), Positives = 117/209 (55%)
 for NOV4: 16 to 222; Sbjct: 116 to 324
Expect = 2.1e-41,Identities = 19/39 (48%), Positives = 28/39 (71%)
for NOV4: 290 to 328; Sbjct: 344 to 382

prdm:29671 p36 (1) PMFF_PROMI // PUTATIVE MINOR FIMBRIAL SUBUNIT PMFF PRECURSOR.
FIMBRIA; SIGNAL, 53 aa.
Expect = 0.64, Identities = 15/48 (31%), Positives = 27/48 (56%)
for NOV4: 157 to 202; Sbjct: 6 to 53

prdm:16833 p36 (2) VL96(2) // L96 PROTEIN REPEAT DNA PACKAGING DNA-BINDING, 61 aa.
Expect = 2.2, Identities = 11/32 (34%), Positives = 18/32 (56%)
for NOV4: 21 to 52; Sbjct: 9 to 40

prdm:2442 p36 (10) INVO(10) // INVOLUCRIN KERATINOCYTE REPEAT, 65 aa.
Expect = 4.7, Identities = 14/40 (35%), Positives = 20/40 (50%)
for NOV4: 242 to 276; Sbjct: 8 to 47

prdm:15830 p36 (2) GLG1(1) GLG2(1) // GLYCOGEN SYNTHESIS INITIATOR PROTEIN
BIOSYNTHESIS GLG1 GLG2, 51 aa.
Expect = 6.0, Identities = 10/23 (43%), Positives = 14/23 (60%)
for NOV4: 254 to 276; Sbjct: 22 to 44

BLOCKS Protein Domain Analysis
AC#          Description                                    Strength  Score
```

```
BL00115R    0 Eukaryotic RNA polymerase II heptapeptide rep      2074   1110
BL00911C    0 Dihydroorotate dehydrogenase proteins.             1314   1050
BL01137D    0 Uncharacterized protein family UPF0006 protei      1297   1048
BL00576B    0 General diffusion Gram-negative porins protei      1391   1047
BL01182C    0 Glycosyl hydrolases family 35  proteins.          1577   1046

ProSite Protein Domain Analysis                          NOV4 aa position
Pattern-ID: ASN_GLYCOSYLATION PS00001 (Interpro)                   69
Pattern-DE: N-glycosylation site
Pattern:    N[^P][ST][^P]

Pattern-ID: CAMP_PHOSPHO_SITE PS00004 (Interpro)                  334
Pattern-DE: cAMP- and cGMP-dependent protein kinase phosphorylation site
Pattern:    [RK]{2}.[ST]

Pattern-ID: PKC_PHOSPHO_SITE PS00005 (Interpro)          12, 148, 301, 305, 322
Pattern-DE: Protein kinase C phosphorylation site
Pattern:    [ST].[RK]

Pattern-ID: CK2_PHOSPHO_SITE PS00006 (Interpro)           54, 142, 151, 171
Pattern-DE: Casein kinase II phosphorylation site
Pattern:    [ST].{2}[DE]

Pattern-ID: MYRISTYL PS00008 (Interpro)                   94, 111, 183, 308
Pattern-DE: N-myristoylation site
Pattern:    G[^EDRKHPFYW].{2}[STAGCN][^P]
```

Other BLAST results include sequences from the Patp database, which is a proprietary database that contains sequences published in patents and patent publications. A BLASTP analysis of the patp database showed that NOV4 has 85 of 209 aa residues (40%) identical to, and 117 of 209 aa residues (55%) positive with, the 385 aa Saccharomyces cerevisiae Lipid metabolism protein encoded by the open reading frame YGR046w (patp:AAB19189, Expect = 1.6e-40). Patp results include those listed in Table 4G.

Table 4G. Patp alignments of NOV4

Sequences producing High-scoring Segment Pairs:	High Score	Smallest Sum Prob. P(N)
patp:AAB19189 Lipid metabolism protein encoded by the open reading frame YGR046w - Saccharomyces cerevisiae, 385 aa.	374	1.6e-40

The disclosed NOV4 nucleic acid encoding a lipid metabolism associated protein-like protein includes the nucleic acid whose sequence is provided in Table 4A, or a fragment thereof. The invention also includes a mutant or variant nucleic acid any of whose bases may be changed from the corresponding base shown in Table 4A while still encoding a protein that maintains its lipid metabolism -like activities and physiological functions, or a fragment of such a nucleic acid. The invention further includes nucleic acids whose sequences are complementary to those just described, including nucleic acid fragments that are complementary to any of the nucleic acids just described. The invention additionally includes nucleic acids or nucleic acid fragments,

or complements thereto, whose structures include chemical modifications. Such modifications include, by way of nonlimiting example, modified bases, and nucleic acids whose sugar phosphate backbones are modified or derivatized. These modifications are carried out at least in part to enhance the chemical stability of the modified nucleic acid, such that they may be used, for example, as antisense binding nucleic acids in therapeutic applications in a subject. In the mutant or variant nucleic acids, and their complements, up to about 45 % percent of the bases may be so changed.

The disclosed NOV4 protein of the invention includes the lipid metabolism -like protein whose sequence is provided in Table 4B. The invention also includes a mutant or variant protein any of whose residues may be changed from the corresponding residue shown in Table 4B while still encoding a protein that maintains its lipid metabolism -like activities and physiological functions, or a functional fragment thereof. In the mutant or variant protein, up to about 58 % percent of the residues may be so changed.

The invention further encompasses antibodies and antibody fragments, such as F_{ab} or $(F_{ab})_2$ that bind immunospecifically to any of the proteins of the invention.

The above defined information for this invention suggests that this lipid metabolism -like protein (NOV4) may function as a member of a v family". Therefore, the NOV4 nucleic acids and proteins identified here may be useful in potential therapeutic applications implicated in (but not limited to) various pathologies and disorders as indicated below. The potential therapeutic applications for this invention include, but are not limited to: cardiovascular disease research tools, for all tissues and cell types composing (but not limited to) those defined here

Based on the tissues in which NOV4 is most highly expressed; including duodenum, small intestine, uterus, thymus, CAEC, liver, breast, lung, kidney; specific uses include developing products for the diagnosis or treatment of a variety of diseases and disorders. Additional disease indications and tissue expression for NOV4 is presented in Example 2.

The NOV4 nucleic acids and proteins of the invention are useful in potential therapeutic applications implicated in cancer including but not limited to heart disease, stroke and/or other pathologies and disorders. For example, a cDNA encoding the lipid metabolism -like protein (NOV4) may be useful in cardiovascular disease therapy, and the lipid metabolism -like protein (NOV4) may be useful when administered to a subject in need thereof. By way of nonlimiting example, the compositions of the present invention will have efficacy for treatment of patients suffering from cardiovascular disease including but not limited to heart disease, hypertension, diabetes, stroke and renal failure. The NOV4 nucleic acid encoding lipid metabolism -like protein, and the lipid metabolism -like protein of the invention, or fragments thereof, may further

be useful in diagnostic applications, wherein the presence or amount of the nucleic acid or the protein are to be assessed.

NOV4 nucleic acids and polypeptides are further useful in the generation of antibodies that bind immuno-specifically to the novel NOV4 substances for use in therapeutic or diagnostic methods. These antibodies may be generated according to methods known in the art, using prediction from hydrophobicity charts, as described in the "Anti-NOVX Antibodies" section below. The disclosed NOV4 protein has multiple hydrophilic regions, each of which can be used as an immunogen. In one embodiment, a contemplated NOV4 epitope is from about amino acids 1 to 20. In another embodiment, a NOV4 epitope is from about amino acids 30 to 55. In additional embodiments, NOV4 epitopes are from about amino acids 60 to 75, from about amino acids 80-95, from about amino acids 120 to 160, from about amino acids 185-290 and from about amino acids 300-337. These novel proteins can be used in assay systems for functional analysis of various human disorders, which will help in understanding of pathology of the disease and development of new drug targets for various disorders.

NOV5

In another embodiment, the novel sequence is NOV5 (alternatively referred to herein as 24SC239), which includes the 983 nucleotide sequence (SEQ ID NO:26) shown in Table 5A. A NOV5 ORF begins with a Kozak consensus ATG initiation codon at nucleotides 66-68 and ends with a TGA codon at nucleotides 551-553. Putative untranslated regions upstream from the initiation codon and downstream from the termination codon are underlined in Table 5A, and the start and stop codons are in bold letters.

Table 5A. NOV5 Nucleotide Sequence (SEQ ID NO:26)

CCGCGGCTGTGTCGTCATACTTGCGGCGCCGACGCCGCCGCTCGCTTGTGAAACTGGAAGGCTGCC**ATG**GCTAGCCCAGC
CGCCTCCTCGGTGCGACCACCGAGGCCCAAGAAAGAGCCGCAGACGCTCGTCATCCCCAAGAATGCGGCGGAGGAGCAG
AAGCTCAAGCTGGAGCGGCTCATGAAGAACCCGGACAAAGCAGTTCCAATTCCAGAGAAAAATGAGTGAATGGGCACCTC
GACCTCCCCCAGAATTTGTCCGAGATGTCATGGGTTCAAGTGCTGGGGCCGGCAGTGGAGAGTTCCACGTGTACAGACA
TCTGCGCCGGAGAGAATATCAGCGACAGGACTACATGGATGCCATGGCTGAGAAGCAAAAATTGGATGCAGAGTTTCAG
AAAAGACTGGAAAAGAATAAAATTGCTGCAGAGGAGCAGACCGCAAAGCGCCGGAAGAAGCGCCAGAAGTTAAAAGAGA
AGAAATTACTGGCAAAGAAGATGAAACTTGAACAGAAGAAACAAGAAGGACCCGGTCAGCCCAAGGAGCAGGGGTCCAG
CAGCTCTGCGGAGGCATCTGGAACAGAGGAGGAGGAAGTGCCCAGTTTCACCATGGGGCGA**TGA**CAATGTTTGCCA
CAGCCTCTGCCTGGAACCTGGCTCGTGCTGTGACCAGAAGGGAAAGGCGGCTGTTTGGCTCTTTCTCCCCCGCAAGGAC
CCGCTGACCCGCTGGATGGAGAGCAAAGGAGACCCCTCCCGAGCCGCTCACAGTCCTGTATTTGGCAGGTTTGGGAGCC
TGAGGGGCCATCTCCCTGACACTCAGAGGCACTGCCTTGCAGACACCATCCGTGCTCCTGGTAAAGGGGGACAGAGAGC
CTCACCTTGCCACATATTTGAACAGTGATGAGTTTGGGGCTGGTTTCTGGGAAGGGAACGTTTATTTAGTAAAGAGCAG
AACACCCTTAAAAAAAAAAAAAAAAAAAAAAAAAAA

The NOV5 protein (SEQ ID NO:27) encoded by SEQ ID NO:26 is 184 amino acids in length and is presented using the one-letter code in Table 5B. The Psort profile for NOV5 predicts that this sequence has no known signal peptide and is likely to be localized at the nucleus with a certainty of 0.9883. In alternative embodiments, a NOV5 polypeptide is located

to the mitochondrial matrix space with a certainty of 0.1000, or the lysosome (lumen) with a certainty of 0.1000. The NOV5 protein has a molecular weight of 20996.9 Daltons.

Table 5B. NOV5 protein sequence (SEQ ID NO:27)

```
MASPAASSVRPPRPKKEPQTLVIPKNAAEEQKLKLERLMKNPDKAVPIPEKMSEWAPRPPPEFVRDVMGSSAGAGSGEF
HVYRHLRREYQRQDYMDAMAEKQKLDAEFQKRLEKNKIAAEEQTAKRRKKRQKLKEKKLLAKKMKLEQKKQEGPGQPK
EQGSSSSAEASGTEEEEEVPSFTMGR
```

The reverse complement for NOV5 is presented in Table 5C.

Table 5C. NOV5 reverse complement (SEQ ID NO:28)

```
TTTTTTTTTTTTTTTTTTTTTTTTTAAGGGTGTTCTGCTCTTTACTAAATAAACGTTCCCTTCCCAGAAACCAGCCCC
AAACTCATCACTGTTCAAATATGTGGCAAGGTGAGGCTCTCTGTCCCCCTTTACCAGGAGCACGGATGGTGTCTGCAAG
GCAGTGCCTCTGAGTCTCAGGCAGATGGCCCCTCAGGCTCCCAAACTTGCCAAATACAGGACTGTGAGCGGCTCGGGAG
GGGTCTCCTTTGCTCTCCATCCAGCGGGTCAGCGGGTCCTTGCGGGGGAGAAAGAGCCAAACAGCCGCCTTTCCCTTCT
GGTCACAGCACGAGCCAGGTTCCAGGCAGAGGCTGTGGCAAACATTGTCATCGCCCCATGGTGAAACTGGGCACTTCCT
CCTCCTCCTCTGTTCCAGATGCCTCCGCAGAGCTGCTGGACCCCTGCTCCTTGGGCTGACCGGGTCCTTCTTGTTTCTT
CTGTTCAAGTTTCATCTTCTTTGCCAGTAATTTCTTCTCTTTTAACTTCTGGCGCTTCTTCCGGCGCTTTGCGGTCTGC
TCCTCTGCAGCAATTTTATTCTTTTCCAGTCTTTTCTGAAACTCTGCATCCAATTTTTGCTTCTCAGCCATGGCATCCA
TGTAGTCCTGTCGCTGATATTCTCTCCGGCGCAGATGTCTGTACACGTGGAACTCTCCACTGCCGGCCCCAGCACTTGA
ACCCATGACATCTCGGACAAATTCTGGGGGAGGTCGAGGTGCCCATTCACTCATTTTCTCTGGAATTGGAACTGCTTTG
TCCGGGTTCTTCATGAGCCGCTCCAGCTTGAGCTTCTGCTCCTCCGCCGCATTCTTGGGGATGACGAGCGTCTGCGGCT
CTTTCTTGGGCCTCGGTGGTCGCACCGAGGAGGCGGCTGGGCTAGCCATGGCAGCCTTCCAGTTTCACAAGCGAGCGGC
GGCGTCGGCGCGCAAGTATGACGACACAGCCGCGG
```

BLASTP results for NOV5 are shown in Table 5D.

Table 5D. BLAST results for NOV5

Matching Entry (in SwissProt + SpTrEMBL)	Description	aa Length	% Identity	% Positive	E Value
Q9H875; AK023964; BAB14742.1	CDNA FLJ13902 FIS, CLONE THYRO1001793. homo sapiens. 3/2001	184	184/184 (100%)	184/184, (100%)	1e-102
Q9CWV6; AK010359; BAB26879.1	8430424D23RIK PROTEIN. mus musculus. 6/2001	186	170/186 (91%)	174/186, (94%)	4e-89
Q9CY32; AK010359; BAB26879.1	8430424D23RIK PROTEIN. mus musculus. 6/2001	186	170/186 (91%)	174/186, (94%)	4e-89
Q9CXA5; AK018438; BAB31212.1	8430424D23RIK PROTEIN. mus musculus. 6/2001	148	133/148 (90%)	136/148, (92%)	2e-67
Q9V7K1; AE003808; AAF58048.1	CG8441 PROTEIN. drosophila melanogaster 5/2000	253	75/158 (47%)	99/158, (63%)	3e-30

A multiple sequence alignment is given in Table 5E, with the NOV5 protein of the invention being shown on lines 1 in a ClustalW analysis comparing NOV5 with related protein sequences of Table 5D.

Table 5E. Information for the ClustalW proteins:

1. SEQ ID NO:27, NOV5
2. SEQ ID NO:29, Q9H875 CDNA FLJ13902 FIS, CLONE THYRO1001793. homo sapiens. 3/2001
3. SEQ ID NO:30, Q9CWV6 8430424D23RIK PROTEIN. mus musculus 6/2001
4. SEQ ID NO:31, Q9CY32 8430424D23RIK PROTEIN. mus musculus 6/2001
5. SEQ ID NO:32, Q9CXA5 8430424D23RIK PROTEIN. mus musculus. 6/2001
6. SEQ ID NO:33, Q9V7K CG8441 PROTEIN. drosophila melanogaster. 5/2000

```
NOV5          1   -------MASP---------AASSVRPPRPKPEPQTLVIPKNAAEPQLKLPELMKNPDK  44
Q9H875        1   -------MASP---------AASSVRPPRPKPEPQTLVIPKNAAEPQLKLPELMKNPDK  44
Q9CWV6        1   -------MASP---------AASSVRPPRPKPEPQTLVIPKNAAEPQLKLPELMKNPDK  44
Q9CY32        1   -------MASP---------AASSVRPPRPKPEPQTLVIPKNAAEPQLKLPELMKNPDK  44
Q9CXA5        1   ------------------------------------------------MKNPDK   6
Q9V7K1        1   MSLIKNLVKEPEQQAKKKKNNAGSESDSDEDQKPLRPFIKPATDLGRLKLPELMKNPDK  60

NOV5         45   AVPIPEKMSEWAPKP-PPEFVRDVMGSSAGAGSGEPHVYRHLRRREYQRQDYMDAMAEKQ  103
Q9H875       45   AVPIPEKMSEWAPKP-PPEFVRDVMGSSAGAGSGEPHVYRHLRRREYQRQDYMDAMAEKQ  103
Q9CWV6       45   AVPIPEKMSEWAPWA-PPEFVRDVMGSSAGAGSGEPHVYRHLRRREYQRQDYMDAMAEKQ  103
Q9CY32       45   AVPIPEKMNEWAPKA-PPEFVRDVMGSSAGAGSGEPHVYRHLRRREYQRQDYMDAMAEKQ  103
Q9CXA5        7   AVPIPEKMSEWAPKA-PPEFVRDVMGSSAGAGSGEPHVYRHLRRREYQRQDYMDAMAEKQ  65
Q9V7K1       61   PTVIPEQRPERDFMSSVPTFVNVMGSSAGAGSGEPHVYRHLRAPEYRQKNIQNQSARE  120

NOV5         104  KLDAEPQKRLEKNKIAAEEQTAKRRKKRQKLP--------EKKLLAKKMKLEQKKQ--EG  153
Q9H875       104  KLDAEPQKRLEKNKIAAEEQTAKRRKKRQKLP--------EKKLLAKKMKLEQKKQ--EG  153
Q9CWV6       104  KLDAEPQKRLEKNKIAAEEQTAKRRKKRQKLP--------EKKLLAKKMKLEQKKQKKEE  154
Q9CY32       104  KLDAEPQKRLEKNKIAAEEQTAKRRKKRQKLP--------EKKLLAKKMKLEQKKQKKEE  154
Q9CXA5        66  KLDAEPQKRLEKNKIAAEEQTAKRRKKRQKLP--------EKKLLAKKMKLEQKKQKKEE  116
Q9V7K1       121  AAEEAVQKLVDERRRAEEETAKRRKKLAKRQRAKKPREVKPLDLKEASEDSNTDSEEE  180

NOV5         154  QQQPKD--------QQSSSQLASQTEEE--EDVPS-PTMGR----------------  184
Q9H875       154  QQQPKD--------QQSSSQLASQTEEE--EDVPS-PTMGR----------------  184
Q9CWV6       155  QQQQQD--------QHSSSQLASQTEEE--EDEPSVLIMGR----------------  186
Q9CY32       155  QQQQQD--------QHSSSQLASQTEEE--EDEPSVVIMGR----------------  186
Q9CXA5       117  QQQQQD--------QHSSSQLASQTEEE--EDEPSVVIMGR----------------  148
Q9V7K1       181  QTEEKAESSPEEGQQVQSKESDDNVQETSNSEDAVQSNTEAESAEDTNAVELDSTEATKE 240

NOV5         184  ------------ 184
Q9H875       184  ------------ 184
Q9CWV6       186  ------------ 186
Q9CY32       186  ------------ 186
Q9CXA5       148  ------------ 148
Q9V7K1       241  SQNVDQEQDKPVP 253
```

ProDom results for NOV5 were collected from using a proprietary database. The results are listed in Table 5F with the statistics and domain description.

Table 5F. ProDom results for NOV5

ProDom Analysis Sequences producing High-scoring Segment Pairs:	High Score	Smallest Sum Probability P(N)
prdm:38062 p36 (1) INCE_CHICK // INNER CENTROMERE PROTEIN...	119	1.1e-06
prdm:26211 p36 (1) D7_DICDI // CAMP-INDUCIBLE PRESSPORE PR...	82	0.00051
prdm:4957 p36 (5) CALD(5) // CALDESMON CDM MUSCLE PROTE...	74	0.0041
prdm:22005 p36 (1) INCE_CHICK // INNER CENTROMERE PROTEIN...	72	0.0070

>prdm:38062 p36 (1) INCE_CHICK // INNER CENTROMERE PROTEIN (INCENP). CELL DIVISION; MICROTUBULES; COILED COIL; CENTROMERE; MITOSIS; CELL CYCLE; NUCLEAR PROTEIN; ALTERNATIVE SPLICING, 218 aa.
Identities = 31/94 (32%), Positives = 57/94 (60%) for NOV5: 86-179, Sbjct: 9-98
Identities = 29/97 (29%), Positives = 55/97 (56%) for NOV5: 86-182, Sbjct: 9-104
Identities = 24/79 (30%), Positives = 46/79 (58%) for NOV5: 98-176, Sbjct: 2-73

```
>prdm.26211 p36 (1) D7_DICDI // CAMP-INDUCIBLE PRESPORE PROTEIN D7 PRECURSOR.
SPORULATION; SIGNAL, 112 aa.
Identities = 24/90 (26%), Positives = 47/90 (52%) for NOV5: 88 -177, Sbjct: 16-96
Identities = 21/76 (27%), Positives = 38/76 (50%) for NOV5: 8-152, Sbjct: 16-91

>prdm:4957 p36 (5) CALD(5) // CALDESMON CDM MUSCLE PROTEIN ACTIN-BINDING CALMODULIN-
BINDING PHOSPHORYLATION ALTERNATIVE SPLICING REPEAT, 89 aa.
Identities = 24/73 (32%), Positives = 40/73 (54%) for NOV5: 11-184, Sbjct: 8-80

>prdm:22005 p36 (1) INCE_CHICK // INNER CENTROMERE PROTEIN (INCENP). CELL DIVISION;
MICROTUBULES; COILED COIL; CENTROMERE; MITOSIS; CELL CYCLE; NUCLEAR PROTEIN;
ALTERNATIVE SPLICING, 71 aa.
Identities = 18/67 (26%), Positives = 40/67 (59%) for NOV5: 96-160, Sbjct: 2-68
Identities = 16/56 (28%), Positives = 29/56 (51%) for NOV5: 86-71, Sbjct: 16-71

PROSITE - Protein Domain Matches for Gene ID: NOV05
Pattern-ID: PKC_PHOSPHO_SITE PS00005 (Interpro) PDOC00005
Pattern-DE: Protein kinase C phosphorylation site
Pattern:    [ST].[RK]

Pattern-ID: CK2_PHOSPHO_SITE PS00006 (Interpro) PDOC00006
Pattern-DE: Casein kinase II phosphorylation site
Pattern:    [ST].{2}[DE]

Pattern-ID: MYRISTYL PS00008 (Interpro) PDOC00008
Pattern-DE: N-myristoylation site
```

The INCE_CHICK // INNER CENTROMERE PROTEIN (INCENP) is involved in cell division, microtubules, and centromeres. It is also involved with cell cycle through involvement with nuclear proteins and alternative splicing. The D7_DICDI // CAMP-INDUCIBLE PRESPORE PROTEIN D7 PRECURSOR is involved with cell signaling and sporulation.

BLOCKS analysis was also performed on NOV5. Protein families that NOV5 was similar to are shown in Table 5G.

Table 5G. BLOCKS Analysis of NOV5

AC#	Description	Strength	Score
BL00500	0 Thymosin beta-4 family proteins.	1993	1089
BL01103E	0 Aspartate-semialdehyde dehydrogenase proteins	1372	1057
BL00936A	0 Ribosomal protein L35 proteins.	1518	1039
BL01002C	0 Translationally controlled tumor protein.	1430	1026
BL01179A	0 Phosphotyrosine interaction domain proteins (	1196	1025
BL01104C	0 Ribosomal protein L13e proteins.	1458	1022
BL00412B	0 Neuromodulin (GAP-43) proteins.	1927	1006
BL01252D	0 Endogenous opioids neuropeptides precursors p	1763	1005
BL01118B	0 Translation initiation factor SUI1 proteins.	1517	1003
BL00892B	0 HIT family proteins.	1500	1002

Other BLAST results include sequences from the Patp database, which is a proprietary database that contains sequences published in patents and patent publications. Patp results include those listed in Table 5H.

Table 5H. Patp alignments of NOV5

Sequences producing High-scoring Segment Pairs:

	% Identity	% Positive
patp:AAB50322 Human cytoskeleton-associated protein #2 - .	100%	100%
patp:AAB94798 Human protein sequence SEQ ID NO:15925 - Ho. .	100%	100%
patp:AAG42902 Arabidopsis thaliana protein fragment SEQ I. .	45%	57%
patp:AAG42903 Arabidopsis thaliana protein fragment SEQ I. .	45%	57%
patp:AAG42904 Arabidopsis thaliana protein fragment SEQ I...	45%	57%
patp:AAG51246 Arabidopsis thaliana protein fragment SEQ I...	47%	58%
patp:AAG51247 Arabidopsis thaliana protein fragment SEQ I...	47%	58%
patp:AAG51248 Arabidopsis thaliana protein fragment SEQ I...	47%	58%

NOV5 is expressed in at least the following tissues: lung, ovary, prostate, tonsil, breast cancer, and ovarian cancer. This information was derived by determining the tissue sources of the sequences that were included in the invention including but not limited to SeqCalling sources, Public EST sources, Literature sources, and/or RACE sources.

The disclosed NOV5 nucleic acid encoding a novel protein includes the nucleic acid whose sequence is provided in Table 5A, or a fragment thereof. The invention also includes a mutant or variant nucleic acid any of whose bases may be changed from the corresponding base shown in Table 5A while still encoding a protein that maintains its activities and physiological functions, or a fragment of such a nucleic acid. The invention further includes nucleic acids whose sequences are complementary to those just described, including nucleic acid fragments that are complementary to any of the nucleic acids just described. The invention additionally includes nucleic acids or nucleic acid fragments, or complements thereto, whose structures include chemical modifications. Such modifications include, by way of nonlimiting example, modified bases, and nucleic acids whose sugar phosphate backbones are modified or derivatized. These modifications are carried out at least in part to enhance the chemical stability of the modified nucleic acid, such that they may be used, for example, as antisense binding nucleic acids in therapeutic applications in a subject. In the mutant or variant nucleic acids, and their complements, up to about 37 % percent of the bases may be so changed.

The disclosed NOV5 protein of the invention includes thenovel protein whose sequence is provided in Table 5B. The invention also includes a mutant or variant protein any of whose residues may be changed from the corresponding residue shown in Table 5B while still encoding a protein that maintains its activities and physiological functions, or a functional fragment thereof. In the mutant or variant protein, up to about 37 % percent of the residues may be so changed.

The invention further encompasses antibodies and antibody fragments, such as F_{ab} or $(F_{ab})_2$ that bind immunospecifically to any of the proteins of the invention.

The NOV5 nucleic acids and proteins of the invention are useful in potential therapeutic applications implicated in cancer including but not limited to breast cancer, ovarian cancer, and/or other pathologies and disorders. For example, a cDNA encoding the novel protein (NOV5) may be useful in cancer therapy, and thenovel protein (NOV5) may be useful when administered to a subject in need thereof. By way of nonlimiting example, the compositions of the present invention will have efficacy for treatment of patients suffering from cancer including but not limited to breast and ovarian cancer. The NOV5 nucleic acid encoding novel protein, of the invention, or fragments thereof, may further be useful in diagnostic applications, wherein the presence or amount of the nucleic acid or the protein are to be assessed.

NOV5 nucleic acids and polypeptides are further useful in the generation of antibodies that bind immuno-specifically to the novel NOV5 substances for use in therapeutic or diagnostic methods. These antibodies may be generated according to methods known in the art, using prediction from hydrophobicity charts, as described in the "Anti-NOVX Antibodies" section below. The disclosed NOV5 protein has multiple hydrophilic regions, each of which can be used as an immunogen. In one embodiment, a contemplated NOV5 epitope is from about amino acids 1 to 20. In another embodiment, a NOV5 epitope is from about amino acids 25 to 45. In additional embodiments, NOV5 epitopes are from about amino acids 50 to 55, from about amino acids 60 to 70, from about amino acids 85 to 100, and from about amino acids 105 to 175. These novel proteins can be used in assay systems for functional analysis of various human disorders, which will help in understanding of pathology of the disease and development of new drug targets for various disorders.

NOV6

In another embodiment, the E1F-2B epsilon subunit-like protein is NOV6 (alternatively referred to herein as 24SC300), which includes the 2456 nucleotide sequence (SEQ ID NO:34) shown in Table 6A. A NOV6 ORF begins with a Kozak consensus ATG initiation codon at nucleotides 836-838 and ends with a TGA codon at nucleotides 1934-1936. Putative untranslated regions upstream from the initiation codon and downstream from the termination codon are underlined in Table 6A, and the start and stop codons are in bold letters.

Table 6A. NOV6 Nucleotide Sequence (SEQ ID NO:34)

GAATTCCTGACTGCCACAGGTGTACAGGAAACATTTGTCTTTTGTTGCTGGAAAGCTGCTCAAATCAAAGAACA
TTTACTGAAGTCAAAGTGGTGCCGCCCTACATCTCTCAATGTGGTTCGAATAATTACATCAGAGCTCTATCGAT
CACTGGGAGATGTCCTCCGTGATGTTGATGCCAAGGCTTTGGTGCGCTCTGACTTTCTTCTGGTGTATGGGGAT
GTCATCTCAAACATCAATATCACCAGAGCCCTTGAGGAACACAGGTTGAGACGGAAGCTAGAAAAAAAATGTTTC
TGTGATGACGATGATCTTCAAGGAGTCATCCCCCAGCCACCCAACTCGTTGCCACGAAGACAATGTGGTAGTGG
CTGTGGATAGTACCACAAACAGGGTTCTCCATTTTCAGAAGACCCAGGGTCTCCGGCGTTTTGCATTTCCTCTG
AGCCTGTTTCAGGGCAGTAGTGATGGAGTGGAGGTTCGATATGATTTACTGGATTGTCATATCAGCATCTGTTC
TCCTCAGGTGGCACAACTCTTTACAGACAACTTTGACTACCAAACTCGAGATGACTTTGTGCGAGGTCTCTTAG
TGAATGAGGAGATCCTAGGGAACCAGATCCACATGCACGTAACAGCTAAGGAATATGGTGCCCGTGTCTCCAAC

CTACACATGTACTCAGCTGTCTGTGCTGACGTCATCCGCCGATGGGTCTACCCTCTCACCCCAGAGGCGAACTT
CACTGACAGCACCACCCAGAGCTGCACTCATTCCCGGCACAACATCTACCGAGGGCCTGAGGTCAGCCTGGGCC
ATGGCAGCATCCTAGAGGAAAATGTGCTCCTGGGCTCTGGCACTGTCATTGGCAGCAATTGCTTTATCACCAAC
AGTGTCATTGGCCCGGCTGCCACATTGGTGAGCACAGGTGATAACCTGGTGCTGGACCAGACTACCTGTGGC
AGGGTGTTCGAGTGGCGGATGGAGCACAGATCCATCAGTCTCTGCCTTGTGACAATGCTGAGGTCAGGAACGA
GTGACCTGAAACCACGCTCTGTCCACTTCCCAGGTGGTCGTGGGCCCAAATATCACGCTGAGGGCTC
GGTGATCTCTTTGCACCCTCCAGATGCAGAGGAAGATGAAGATGATGGCGAGTTCAGTGATGATTCTGGGGCTG
ACCAAGAAAAGGACAAAGTGAAGATGAAAGGTTACAATCCAGCAGAAGTAGGAGCTGCTGGCAAGGGCTACCTC
TGGAAAGCTGCAGGCATGAACATGGAGGAAGAGGAGGAACTGCAGCAGAATCTGTGGGGACTCAAGATCAACAT
GGAAGAAGGAGTGAAAGTGAAAGTGAGCAAAGTATGGATTCTGAGGAGCCGGACAGCCGGGGAGGCTCCCCTC
AGATGGATGACATCAAAGTGTTCCAGAATGAAGTTTAGGAACACTACAGCGGGGCAAAGAGGAGAACATTTCT
TGTGACAATCTCGTCCTGGAAATCAACTCTCTCAAGTATGCCTATAACATAAGTCTAAAGAGGTGATGCAGGT
ACTGAGCCACGTGGTCCTGGAGTTCCCCCTGCAACAGATGGATTCCCCGCTTGACTCAAGCCGCTACTGTGCCC
TGCTGCTTCCTCTGCTAAAGGCCTGGAGCCCTGTTTTTAGGAACTACATAAAGCGCGCAGCCGACCATTTGGAA
GCGTTAGCAGCCATTGAGGACTTCTTCCTAGAGCATGAAGCTCTTGGTATTTCCATGGCCAAGGTACTGATGGC
TTTCTACCAGCTGGAGATCCTGGCTGAGGAAACAATTCTGAGCTGGTTCAGCCAAAGAGATACAACTGACAAGG
GCCAGCAGTTGCGCAAGAATCAACAGCTGCAGAGGTTCATCCAGTGGCTAAAAGAGGCAGAAGAGGAGTCATCT
GAAGATGACTGAAGTCACACTGCCTGCTCTTTGGGTGTGATTGAGTCCCTCCTGGCTCCTGGCTCCTGGGCTGGACAA
GTGAGGACTAGCTGCAGAGGATGAGTGACCACCATCCAGGCTGAGACTGAAAGGAGCAGAGGCTGGAACTAC
AGTATTCTTTCCCTCCTAGCAACCATGTGCCTCCCATCCTGACTGTGGAGTTGGGATGTGGAAGTGGGCTGG
AACAAAGCTTCTGCCTAGGGAGGAGCTAAGCAGGCCCGGCAGTTGGAGGAAGGCCAGAGGAACAGCTTTGTGCT
CCGGCTTTCCCTCAGGGAACAGCAGAGAGCAGTTGGCTCTTTCTGCTGCTTGTATATGTTAATATTAAAAGAGA
GAGTGGTGTATTTGGTTTGTCTCCATCCCCGACTAATCAGCCAGTGAAGTATGTGACCAGAATCACATGATAGC
CTTTCCTTAACACCTGGGGGAGAGGGAGGACGGGTGTGCCAGCCACTAGGTGGTACTGTGGTACCTTGCTAATT
AACCTTTCCCATGG

The NOV6 40789.4 Dalton protein (SEQ ID NO:35) encoded by SEQ ID NO:34 is 366 amino acids in length and is presented using the one-letter code in Table 6B. The Psort profile for NOV6 predicts that this sequence has a signal peptide. The most likely cleavage site for a NOV6 peptide is between amino acids 21-22, *i.e.* at the dash between amino acids VSL-AP. NOV6 is likely to be localized outside the cell with a certainty of 0.6138. In alternative embodiments, a NOV6 polypeptide is located to the lysosome (lumen) with a certainty of 0.01900, the endoplasmic reticulum (membrane) with a certainty of 0.1000, or the endoplasmic reticulum(lumen) with a certainty of 0.1000.

Table 6B. NOV6 protein sequence (SEQ ID NO:35)

MCSWALALSLAAIALSPTVSLAPAATLVSTGDNVVLDQTYLWQGVRVAAGAQIHQSLLCDNAEVKERVTLKPRSVLTS
QVVVGPNITLPEGSVISLHPPDAEEDEDDGEFSDDSGADQEKDKVKMKGYNPAEVGAAGKGYLWKAAGMNMEEEEELQ
QNLWGLKINMEEESESEQSMDSEEPDSRGGSPQMDDIKVFQNEVLGTLQRGKEENISCDNLVLEINSLKYAYNISL
KEVMQVLSHVVLEPPLQQMDSPLDSSRYCALLLPLLKAWSPVFRNYIKRAADHLEALAAIEDPFLEHEALGISMAKVL
MAFYQLEILAEETILSWFSQRDTTDRGQQLKRNQQLQRFIQWLKEAEEESEDD

The reverse complement for NOV6 is presented in Table 6C.

Table 6C. NOV6 reverse complement (SEQ ID NO:36)

CCATGGGAAAGGTTAATTAGCAAGGTACCACAGTACCACCTAGTGGCTGGCACACCCGTCCTCCCTCTCCCCCAGGTG
TTAAGGAAAAGGCTATCATGTGATTCTGGTCACATACTTCACTGGCTGATTAGTCGGGGATGGAGACAAACCAAATACA
CCACTCTCCTCTTTAATATTACATATACAAGCAGCAGAAAGAGCCAACTGCTCTCTGCTGTTCCCTGAGGGAAAGCC
GGAGCACAAAGCTGTTCCTCTGGCCTTCCTCCAACTGCCGGGCCTGCTTAGCTCCTCCCTAGGCAGAAGCTTTGTTCC
AGCCCCACTTCCACATCCCAACTCCACAGTCAGGATGGGAGGCACATGGTTGCTAGCAGGGGAAAGAATACTGTAGTT
CCAGCCTCTGCTCCTTTCAGTCTCAGCCTGGATGGTGGTCACTCATCCCTCTGCAGCTAGTTCCTCACTTGTCCCAGC
CCAGGAGCCAGGAGGGCACTCAATCACACCCAAAGGAGCAGGCAGTGTGACTTCAGTCATCTTCAGATGACTCCTCTT
CTGCCTCTTTTAGCCACTGGATGAACCTCTGCAGCTGTTGATTCTTGCGCAACTGCTGGCCCTTGTCAGTTGTATCTC
TTTGGCTGAACCAGCTCAGAATTGTTTCCTCAGCCAGGATCTCCAGGCTGGTAGAAAGCCATCAGTACCTTGGCCATGG
AAATACCAAGAGCTTCATGCTCTAGGAAGAAGTCCTCAATGGCTGCTAACGCTTCCAAATGGTCGGCTGCGCGCTTTA
TGTAGTTCCTAAAAACAGGGCTCCAGGCCTTTAGCAGAGGAAGCAGCAGGGCACAGTAGCGGCTTGAGTCAAGCGGG

AATCCATCTGTTGCAGGGGGAACTCCAGGACCACGTGGCTCAGTACCTGCATCACCTCCTTTAGACTTATGTTATAGG
CATACTTGAGAGAGTTGATTTCCAGGACGAGATTGTCACAAGAAATGTTCTCCTCTTTGCCCCGCTGTAGTGTTCCTA
AAACTTCATTCTGGAACACTTTGATGTCATCCATCTGAGGGGAGCCTCCCCGGCTGTCCGGCTCCTCAGAATCCATAC
TTTGCTCACTTTCACTTTCACTCTCTTCTTCCATGTTGATCTTGAGTCCCCACAGATTCTGCTGCAGTTCCTCCTCTT
CCTCCATGTTCATGCCTGCAGCTTTCCAGAGGTAGCCCTTGCCAGCAGCTCCTACTTCTGCTGGATTGTAACCTTTCA
TCTTCACTTTGTCCTTTTCTTGGTCAGCCCCAGAATCATCACTGAACTCGCCATCATCTTCATCTTCCTCTGCATCTG
GAGGGTGCAAAGAGATCACCGAGCCCTCAGGCAGCGTGATATTTGGGCCCACGACCACCTGGGAAGTGAGGACAGAGC
GTGGTTTCAGTGTCACTCGTTCCTTGACCTCAGCATTGTCACAAAGCAGAGACTGATGGATCTGTGCTCCAGCCGCCA
CTCGAACACCCTGCCACAGGTAGGTCTGGTCCAGCACCACGTTATCACCTGTGCTCACCAATGTGGCAGCCGGGGCCA
ATGACACTGTTGGTGATAAAGCAATTGCTGCCAATGACAGTGCCAGAGCCCAGGAGCACATTTTCCTCTAGGATGCTG
CCATGGCCCAGGCTGACCTCAGGCCCTCGGTAGATGTTGTGCCGGGAATGAGTGCAGCTCTGGGTGGTGCTGTCAGTG
AAGTTCGCCTCTGGGGTGAGAGGGTAGACCCATCGGCGGATGACGTCAGCACAGACAGCTGAGTACATGTGTAGGTTG
GAGACACGGGCACCATATTCCTTAGCTGTTACGTGCATGTGGATCTGGTTCCCTAGGATCTCCTCATTCACTAAGAGA
CCTCGCACAAAGTCATCTCGAGTTTGGTAGTCAAAGTTGTCTGTAAAGAGTTGTGCCACCTGAGGAGAACAGATGCTG
ATATGACAATCCAGTAAATCATATCGAACCTCCACTCCATCACTACTGCCCTGAAACAGGCTCAGAGGAAATGCAAAA
CGCCGGAGACCCTGGGTCTTCTGAAAATGGAGAACCCTGTTTGTGGTACTATCCACAGCCACTACCACATTGTCTTCG
TGGCAACGAGTTGGGTGGCTGGGGGATGACTCCTTGAAGATCATCGTCATCACAGAAACATTTTTTTCTAGCTTCCGT
CTCAACCTGTGTTCCTCAAGGGCTCTGGTGATATTGATGTTTGAGATGACATCCCCATACACCAGAAGAAAGTCAGAG
CGCACCAAAGCCTTGGCATCAACATCACGGAGGACATCTCCCAGTGATCGATAGAGCTCTGATGTAATTATTCGAACC
ACATTGAGGATCTAGGCCGCACCACTTTGACTTCAGTAAATGTTCTTTGATTTGAGCAGCTTTCCAGCAACAAAAG
ACAAATGTTTCCTGTACACCTGTGGCAGTCAGGAATTC

BLASTP results for NOV6 are shown in Table 6D.

Table 6D. BLAST results for NOV6

Matching Entry (in SwissProt + SpTrEMBL)	Description	aa Length	% Identity	% Positive	E Value
E2BE_HUMAN; U23028; AAC50646.1	TRANSLATION INITIATION FACTOR EIF-2B EPSILON SUBUNIT (EIF-2B GDP-GTPEXCHANGE FACTOR) (FRAGMENT). homo sapiens. 7/1999	641	335/336 (100%)	336/336, (100%)	0.0
E2BE_RABIT; U23037; AAC48618.1	TRANSLATION INITIATION FACTOR EIF-2B EPSILON SUBUNIT (EIF-2B GDP-GTPEXCHANGE FACTOR). oryctolagus cuniculus. 7/1999	721	294/336 (88%)	318/336, (95%)	1e-171
E2BE_RAT; U19516; AAB17690.1	TRANSLATION INITIATION FACTOR EIF-2B EPSILON SUBUNIT (EIF-2B GDP-GTPEXCHANGE FACTOR). rattus norvegicus. 7/1999	716	292/336 (87%)	314/336, (93%)	1e-168
O64760; AC004238; AAC12836.1	PUTATIVE TRANSLATION INITIATION FACTOR EIF-2B-EPSILON SUBUNIT. arabidopsis thaliana. 6/2001	730	100/362 (28%)	170/362, (47%)	1e-34
Q9SRU3; AC009755; AAF02111.1	PUTATIVE TRANSLATION INITIATION FACTOR EIF-2B EPSILON SUBUNIT. arabidopsis thaliana. 6/2001	676	96/341 (28%)	166/341, (49%)	8e-29

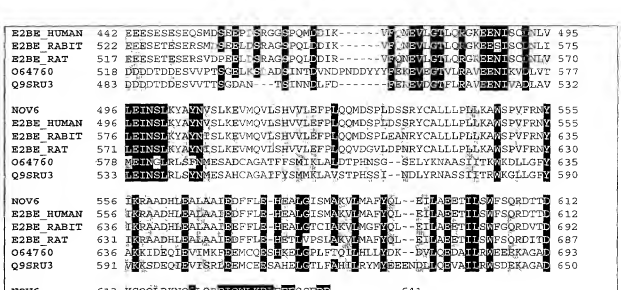
A multiple sequence alignment is given in Table 6E, with the NOV6 protein of the invention being shown on lines 1 in a ClustalW analysis comparing NOV6 with related protein sequences of Table 6D.

Table 6E. Information for the ClustalW proteins:

1. SEQ ID NO:35, NOV6

2. SEQ ID NO:37, E2BE_HUMAN EIF-2B GDP-GTPEXCHANGE FACTOR 7/1999
3. SEQ ID NO:38, E2BE_RABIT EIF-2B GDP-GTPEXCHANGE FACTOR 7/1999
4. SEQ ID NO:39, E2BE_RAT EIF-2B GDP-GTPEXCHANGE FACTOR 7/1999
5. SEQ ID NO:40, O64760 PUTATIVE EIF-2B-EPSILON SUBUNIT 6/2001
6. SEQ ID NO:41, Q9SRU3 PUTATIVE EIF-2B EPSILON SUBUNIT 6/2001

```
NOV6          1  ------------------------------------------------------------  1
E2BE_HUMAN    1  ------------------------------------------------------------  1
E2BE_RABIT    1  MATTVVAPPGAVSDRANKRGGGPGGGGGGGGARGAREESPPPLQAVLVADSPNRRFFPIS 60
E2BE_RAT      1  -MAATAAVPSAVGGRANKRGGGSGGGG----TQGAREEPPPLQAVLVADSPDRFFPIS 55
O64760        1  ------MGAQKKGGAAARVSEDAEVQS-----------RHRLQAILLADSFATKFRPVT 42
Q9SRU3        1  ------MASRKK--RAAKISEDSEEEQS----------RRQRLQAILLADSFATKLLPLT 42

NOV6          1  ----------------------EPLTATGVQETPVFCCWKAACIKGTLKSWVCRPTSLNVV 40
E2BE_HUMAN    1  ----------------------EPLTATGVQETPVFCCWKAACIKGTLKSWVCRPTSLNVV 40
E2BE_RABIT   61  KDQPRVLLPLANVALIDYTLEPLTATGVQETPVFCCWKAACIKGTLKSWVCRPTSLNVV 120
E2BE_RAT     56  KDQPRVLLPLANVALIDYTLEPLTATGVQETPVFCCWKAACIKGTLKSWVCHPTSLNVV 115
O64760       43  LERPKVLLPIVNVPMIDYTLAPESACPEVVPVFCCAHRWQTTSIPSHPNEWVWSHKDFTVK 102
Q9SRU3       43  LERPNVLLPLVNIPMIDYTLAMLESACPEVVPVFC---SWQVIDLPNQSWVWSHKDFTVK 99

NOV6         41  RHITSELYRSIGLVLK----DVDAKALVRSLFNLVVGLVTSNINITRALEDHRRDRKKLEK 96
E2BE_HUMAN   41  RHITSELYRSIGLVLK----DVDAKALVRSLFNLVVGLVTSNINITRALEDHRRDRKKLEK 96
E2BE_RABIT  121  RHITSELYRSIGLVLK----DVDAKALVRSTFNLVVGLVTSNINVTRALEDHRRDRKKLEK 176
E2BE_RAT    116  RHITSDLYRSIGLVLK----DVDAKALVRSLFNLVVGLVTSNINISKALEDHRRDRKKLEK 171
O64760      103  TLES-HKSISGRALNYMYEQQTETSQIQGTPVLVSGDTVSNMPLADLIGEHRRDRKKKDE 161
Q9SRU3      100  THESPQNSTSGRALNYIYEQQIETSQIQGTPVLVNGQIVSNMPLTQLIGEHRRDRKKDE 159

NOV6         97  NVSVMTMVIRKGSSPSL---PDTSCHESNVVVAVLSTSNRVLHFQKTQG---LRRFASPLSL 150
E2BE_HUMAN   97  NVSVMTMVIRKGSSPSL---PDTSCHESNVVVAVLSTSNRVLHFQKTQG---LRRFASPLSL 150
E2BE_RABIT  177  NVSVMTMVIRKGSSPSL---PDTSCHESNVVVAVLSTSNRILHFQKTQG---LRRFSSPLSL 230
E2BE_RAT    172  NVSVMTMVIRKGSSPSL---PDTSCHESNVVLAVLSTSNRILHFQKTQG---LRHFSSPLSL 225
O64760      161  -KAIMTMVIKQSKSSPLTPQSGLGTQQLFIAVDPLAKQELHYEEDKIDHPSGSVCLEKSL 220
Q9SRU3      159  -KAIMTMVIR--------QSLITDHQLFIAVNPLIKQLLWYDRD-------NIGGDKSL 202

NOV6        151  FQGSSSDGVEVRYQLLCQISICSPQVAQLFTLNFDYQTRDDFVRGLLVNEHLCNQHMHS 210
E2BE_HUMAN  151  FQGSSSDGVEVRYQLLCQISICSPQVAQLFTLNFDYQTRDDFVRGLLVNEHLCNQHMHS 210
E2BE_RABIT  231  FQGSGAGVEIRYQLLCQISICSPQVAQLFTLNFDYQTRDDFVRGLLVNEHLCNQHMHS 290
E2BE_RAT    226  FQGSLDGVEIRYQLLCQISICSPQVAQLFTLNFDYQTRDDFVRGLLVNEHLCNQHLHL 285
O64760      221  LD-TNPSVLVCNDQSCYLICSPQVLSLFSLNFDYQSLRHFVKGVLVDDHMCYKLFTH 279
Q9SRU3      203  LD-RNPSVLLCSDQLCYLICSLEVLSLFVLNFDYQIMRCDFVEGVLADDLHCYKLFTH 261

NOV6        211  VTAKEVGARVSNLHMYSAVCADVHSRWVYPLTPEANPTDSTTQSCTHSRHNIVRGPEVSL 270
E2BE_HUMAN  211  VTAKEVGARVSNLHMYSAVCADVHSRWVYPLTPEANPTDSTTQSCTHSRHNIVRGPEVSL 270
E2BE_RABIT  291  VTTREVGARVSNLHMYSAVCADVHSRWVYPLTPEANPTDSTAQSCTHSRHNIVRGPEVSL 350
E2BE_RAT    286  VTSREVGSRVSNLHMYSAVCADVHSRWVYPLTPEVNPTDSSTQSYTHSRHNIVRGPEVSL 345
O64760      280  EIHSSVAGEDNFRSVDTVSKQTLQRWVYPVVPINFSG--NRPLKLGRQGYKASDAVQ 337
Q9SRU3      262  EISSGVASQLPNFRSVDMVSKQTLQRRTFQYVPDMKFSG--NRTLKLERQGYKASDATQ 319

NOV6        271  GHGSILEENVLLCSGTVIGSNCPTNSVIGPGCHIGDNVVLDQTVLWQGVRVAACAQIHQ 330
E2BE_HUMAN  271  GHGSILEENVLLCSGTVIGSNCHTNSVIGPGCHIGDNVVLDQTVLWQGVRVAACAQIHQ 330
E2BE_RABIT  351  GHGSILEENVLLCSGTVIGSNCSTNSVIGPCCIGDNVVLDRAVLWQGVRVAASCAQIHQ 410
E2BE_RAT    346  GHGSVLEENVLLCSGTVIGSNCPTNSVIGPNCHIGDNVVLDQAVLWQGVRVAACAQIHQ 405
O64760      338  SRSADVGASIVLCYGLVLIGDKVLIGSNVVIESSVLMNVTEGSVLINYIEESVLAKNSR 397
Q9SRU3      320  LPSAHVGASIVLCHALVLIGSGTRIGSNVVIGNGCSIGSNVVIQGSVLMNNVVEDGCEIR 379

NOV6        331  SLLCNAEVKERVTLKERSVLTSQVVVCPNITLPEGSVVSLHPPDAEELDDDGEFSPDSG 390
E2BE_HUMAN  331  SLLCNAEVKERVTLKERSVLTSQVVVCPNITLPEGSVVSLHPPDAEELDDDGEFSPDSG 390
E2BE_RABIT  411  SLLCDHAEVKEQVTLKEHSVLTSQVVVCPNITLPEGSVVSLHPPDAEELDDDGQFSPDSG 470
E2BE_RAT    406  SLLCRAEVKERVILKEHSVLTSQVVVCPDIILPEGSVISLHPPDAEELDDDGQFSPDSG 465
O64760      398  AILCHGVKIRAGAVLQEGVVISFNVVVGRDFVVPAYSKVSLLQQPTTLDDELEYAPSS 457
Q9SRU3      380  AIVCIEVKVCAGAIVKEGVVISFNVVVGRDFVVPAYSQVSLLRQPMEELDDE-----E-- 432

NOV6        390  ---ADQEKDKVMKGYNPAEVCAAFKGYLWK------AAGMNMEEEEELQQNLWGLKINM 441
E2BE_HUMAN  390  ---ADQEKDKVKMKGYNPAEVCAAFKGYLWK------AAGMNMEEEEELQQNLWGLKINM 441
O64760      470  ---VNQAKEKAKLKGYNPAEVCVAFKGYLWK------AADMNTEKEEELKQSLWGLTINE 521
E2BE_RAT    465  ---ADQEKEKVKLKGYNPAEVCPEFKGYLWK------AEDVDEKEDEELRQSLWGLMINM 516
O64760      458  SGTADHLSGLNLQMESKASEVCPDFKGYLWGVCEGAHDEEWKHSVAPIPKDKLSEITQAI 517
Q9SRU3      432  ----NLLSGVDLQMESK---IGLDFKGYLWR---QACEDEWKHSVPPIPKDKLAEIIKAI 482

NOV6        442  EEESESSESSEQSMDSEPPSRGGSPQMKDIK------VQQNEVLSTLQRGKEENISCENLV 495
```



ProDom results for NOV6 were collected from a public database. DOMAIN results for NOV6 were collected using the PFAM HMM database. The results are listed in Table 6F with the statistics and domain description.

Table 6F. Domain results for NOV6

ProDom Analysis
prdm:15525 p36 (2) E2BE(2) // TRANSLATION FACTOR EIF-2B INITIATION EPSILON SUBUNIT GDP-GTP EXCHANGE AMINO-ACID BIOSYNTHESIS, 311 aa.
Identities = 270/311 (86%), Positives = 290/311 (93%) for Query: 56-366 and Sbjct: 1-311

>prdm:14746 p36 (2) E2BE(2) // FACTOR TRANSLATION EIF-2B SUBUNIT EXCHANGE INITIATION EPSILON GDP-GTP AMINO-ACID BIOSYNTHESIS, 261 aa.
Identities = 61/245 (24%), Positives = 109/245 (44%) for Query: 129-358 and Sbjct: 17-261

>prdm:3752 p36 (7) IF5(7) // INITIATION FACTOR PROTEIN EUKARYOTIC TRANSLATION EIF-5 BIOSYNTHESIS GTP-BINDING PROBABLE ALTERNATIVE, 260 aa.
Identities = 37/94 (39%), Positives = 51/94 (54%) for Query: 278-363 and Sbjct: 126-219

>prdm:48803 p36 (1) SSRP_DROME // SINGLE-STRAND RECOGNITION PROTEIN (SSRP) (CHORION-FACTOR 5). DNA-BINDING; RNA-BINDING; NUCLEAR PROTEIN, 58 aa.
Identities = 9/20 (45%), Positives = 15/20 (75%) for Query: 100-119 and Sbjct: 2 -20
Identities = 10/29 (34%), Positives = 15/29 (51%) for Query: 165-193 and Sbjct: 29-56

>prdm:25633 p36 (1) FKB1_DROME // 39 KD FK506-BINDING NUCLEAR PROTEIN (PEPTIDYL-PROLYL CIS-TRANS ISOMERASE) (PPIASE) (EC 5.2.1.8). ISOMERASE. ROTAMASE; NUCLEAR PROTEIN, 85 aa
Identities = 27/85 (31%), Positives = 42/85 (49%), for Query: 102-186, Sbjct: 3-78

PFAM HMM Domain Analysis of NOV06

Model	Description	Score	E-value
W2 (InterPro)	eIF4-gamma/eIF5/eIF2-epsilon	121.5	1.6e-32
hormone2 (InterPro)	Peptide hormone	10.4	0.76

Parsed for domains:

Model	Domain	seq-f	seq-t	hmm-f	hmm-t	score	E-value

hormone2	1/1	342	357 ..	13	28 .]	10.4	0	76
W2	1/1	284	366 .]	1	87 []	121.5	1	6e-32

PROSITE - Protein Domain Matches for Gene ID: NOV6

Pattern-ID: ASN_GLYCOSYLATION PS00001 (Interpro) PDOC00001
Pattern-DE: N-glycosylation sites
Pattern: N[^P][ST][^P]
NOV6 Position: 85- NITL; 213-NISC; 231-NISL

Pattern-ID: PKC_PHOSPHO_SITE PS00005 (Interpro) PDOC00005
Pattern-DE: Protein kinase C phosphorylation sites
Pattern: [ST].[RK]
NOV6 Position: 69 -TLK; 225 -SLK; 233-SLK; 259-SSR; 331-SQR; 336-TDK

Pattern-ID: CK2_PHOSPHO_SITE PS00006 (Interpro) PDOC00006
Pattern-DE: Casein kinase II phosphorylation sites
Pattern: [ST].{2}[DE]
NOV6 Position: 29-STGD; 87-TLPE; 114-SGAD; 170-SESE; 233-SLKE; 255-SPLD, 331-SQRD; 362-SSED

Pattern-ID: MYRISTYL PS00008 (Interpro) PDOC00008
Pattern-DE: N-myristoylation sites
Pattern: G[^EDRKHPFYW].{2}[STAGCN][^P]
NOV6 Position: 44-GVRVAA; 91-GSVISL; 161-GLKINM; 305-GISMAK

BLOCKS Analysis

AC#		Description	Strength	Score
BL00260	0	Glucagon / GIP / secretin / VIP family protei	1460	1100
BL00501B	0	Signal peptidases I serine proteins.	1234	1061
BL00558A	0	Eukaryotic mitochondrial porin proteins.	1284	1056
BL00486C	0	DNA mismatch repair proteins mutS family prot	1682	1037
BL00808J	0	ADP-glucose pyrophosphorylase proteins.	1397	1036
BL00992B	0	Serum amyloid A proteins.	1851	1024
BL01271B	0	Sodium:sulfate symporter family proteins.	1480	1022
BL00132E	0	Zinc carboxypeptidases, zinc-binding region 1	1608	1020

The translation factor eif-2B initiation epsilon subunit is involved with GDP-GTP exchange, and amino acid biosynthesis. The initiation factor protein eukaryotic translation EIF-5 is thought to be involved with biosynthesis and GTP-binding. The single-strand recognition protein (SSRP) (chorion-factor 5) is involved with DNA-binding; and RNA- binding. The FK506-binding nuclear protein (peptidyl-prolyl cis-trans isomerase) (PPIASE) (EC 5.2.1.8) is a rotamase; and is involved with nuclear proteins.

Other BLAST results include sequences from the Patp database, which is a proprietary database that contains sequences published in patents and patent publications. Patp results include those listed in Table 6G.

Table 6G. Patp alignments of NOV6

Sequences producing High-scoring Segment Pairs:		
	% Identity	% Positive
patp:AAB43883 Human cancer associated protein sequence SEQ ID NO:1328 - Homo sapiens, 424 aa. PN=WO200055350-A1. Expect = 7.6e-06	29/96 (30%)	55/96 (57%)

The eIF4-gamma/eIF5/eIF2-epsilon proteins are involved with regulation of genes at the translational level, and are involved with GTP-GDP exchange. Peptide hormones are involved in many physiological processes including glucose and fat metabolism, immune system regulation, and neuronal regulation.

NOV6 is expressed in at least the following tissues: placenta, small intestine. larynx, kidney, muscle, colon, tonsil, stomach. uterus, bone marrow, brain and others This information was derived by determining the tissue sources of the sequences that were included in the invention including but not limited to SeqCalling sources, Public EST sources, Literature sources, and/or RACE sources.

The disclosed NOV6 nucleic acid encoding a novel protein includes the nucleic acid whose sequence is provided in Table 6A, or a fragment thereof. The invention also includes a mutant or variant nucleic acid any of whose bases may be changed from the corresponding base shown in Table 6A while still encoding a protein that maintains its activities and physiological functions, or a fragment of such a nucleic acid. The invention further includes nucleic acids whose sequences are complementary to those just described, including nucleic acid fragments that are complementary to any of the nucleic acids just described. The invention additionally includes nucleic acids or nucleic acid fragments, or complements thereto, whose structures include chemical modifications. Such modifications include, by way of nonlimiting example, modified bases, and nucleic acids whose sugar phosphate backbones are modified or derivatized. These modifications are carried out at least in part to enhance the chemical stability of the modified nucleic acid, such that they may be used, for example, as antisense binding nucleic acids in therapeutic applications in a subject. In the mutant or variant nucleic acids, and their complements, up to about 13 % percent of the bases may be so changed.

The disclosed NOV6 protein of the invention includes thenovel protein whose sequence is provided in Table 6B. The invention also includes a mutant or variant protein any of whose residues may be changed from the corresponding residue shown in Table 6B while still encoding a protein that maintains its activities and physiological functions, or a functional fragment thereof. In the mutant or variant protein, up to about 13 % percent of the residues may be so changed.

The invention further encompasses antibodies and antibody fragments, such as F_{ab} or $(F_{ab})_2$ that bind immunospecifically to any of the proteins of the invention.

The NOV6 nucleic acids and proteins of the invention are useful in potential therapeutic applications implicated in cancer including but not limited to breast cancer, ovarian cancer, and/or other pathologies and disorders. For example, a cDNA encoding the novel protein (NOV6) may be useful in cancer therapy, and the novel protein (NOV6) may be useful when

administered to a subject in need thereof. By way of nonlimiting example, the compositions of the present invention will have efficacy for treatment of patients suffering from cancer including but not limited to breast and ovarian cancer. The NOV6 nucleic acid encoding novel protein, of the invention, or fragments thereof, may further be useful in diagnostic applications, wherein the presence or amount of the nucleic acid or the protein are to be assessed.

NOV6 nucleic acids and polypeptides are further useful in the generation of antibodies that bind immuno-specifically to the novel NOV6 substances for use in therapeutic or diagnostic methods. These antibodies may be generated according to methods known in the art, using prediction from hydrophobicity charts, as described in the "Anti-NOVX Antibodies" section below. The disclosed NOV6 protein has multiple hydrophilic regions, each of which can be used as an immunogen. In one embodiment, a contemplated NOV6 epitope is from about amino acids 60 to 75. In another embodiment, a NOV6 epitope is from about amino acids 100 to 135. In additional embodiments, NOV6 epitopes are from about amino acids 145 to 155, from about amino acids 160 to 190, from about amino acids 200 to 220, from about amino acids 230 to 235, from about amino acids 250 to 270, from about amino acids 280 to 290, and from about amino acids 320 to 360. These novel proteins can be used in assay systems for functional analysis of various human disorders, which will help in understanding of pathology of the disease and development of new drug targets for various disorders.

NOV7

In another embodiment, the novel sequence is NOV7 (alternatively referred to herein as 24SC526), which includes the 2004 nucleotide sequence (SEQ ID NO:42) shown in Table 7A. A NOV7 ORF begins with a Kozak consensus ATG initiation codon at nucleotides 176-178 and ends with a TGA codon at nucleotides 404-406. Putative untranslated regions upstream from the initiation codon and downstream from the termination codon are underlined in Table 7A, and the start and stop codons are in bold letters.

Table 7A. NOV7 Nucleotide Sequence (SEQ ID NO:42)

GGGCGCGCGGCCTCGAGGCCTTCCGGTGCGGGAGAAACTACTACTCCCATAATGCCCCGCGGTCCCGCGAGCTG
CCAGTCTCGTCGCGAGAAGCAGCGGCCCGGGGCGACTGAGCGGACAAACGGAAGTGTAGGTTACGGTCTGAGAC
ATCACCGCCAAGCTGGGCATCGGGGAG**ATG**GCCGAGACTGACCCCAAGACCGTGCAGGACCTCACCTCGGTGGT
GCAGACACTCCTGCAGCAGATGCAAGATAAATTTCAGACCATGTCTGACCAGATCATTGGGAGAATTGATGATA
TGAGTAGTCGCATTGATGATCTGGAAAAGAATATCGCGGACCTCATGACACAGGCTGGGGTGGAAGAACTGGAA
AGTGAAAACAAGATACCTGCCACGCAAAAGAGT**TGA**AGGTTGCTAATAATTTATACTGGAATCTGGCATTTTTC
CAAGCCAAGAGAAGATCGAATGGCTTTTTGCAGCTAACTACTATGTGTAGACAGGTTTTATATTATAAAGTATG
CATTCTTATCACCTAGTATATAGTTAGTTTGTAGAGTGATTTCCCCCCAGTTTCTTGAACATGGTATCTTCACA
TCTTGGACCTTGGTCAGTTGTGCTATTCATTATTAAACACTAAAACTTTGGCGGTTCTTGCATAACATTGTCAG
ATTTTTTAGTGTATTTCTGTGAAGTCATTTTTTTCTTGTCATTCCTTTTGTAGTAGTTGCTGTTTGGATAAAA
GTTGATGTGTGATTTTTTATTAAACAAATAGTAAACCCTTCAATTATAGTTAGTCTTGGTGAAGTAAGATGTTT
GTAGACTTTAGAGTTCTTTAATTCTTGGCACAACGTGACTTTTGAGCTAACACCAAATAGTGTGTTGGCAATAC
TTTTCAAATGGCTGAAAACACCTAAAAATTGTTCATTCAGAAATATCTGTCACTGCTCTGTTGCCAAAACTCAG
AATAGAACTTAGACGTATGTCTGAGTCCCTGAGATCACATGCTAAAGTCGATGAAAAGTAACCACTGCCACTGT
CTTGTGTCAGAACTTTTACAGTACAGAAAATAACAGAATAGCCTTCTGTAATGAGGCGTTTGTTAGAGTTTTGC

ATGAGATTCTAATACTTCAGTAGGACCCTACCTACGTGGTTCATCTACAATGGTTACCATAAAAAATCTGGCAG
GATTTTAAAACTCAATCAGTCTTTCCTTTGAGCTAGTGACTTGAAAAGAAGAGAGAAGGAAAAGAGACCATAT
TAAGTCCATGCCAGTTGCTTGGCTAGAATATGATCAACGACTTGTAGTAGACTCAAGTTTTAAAAAACACTAT
TTTACTAAACTGTTTCTTATCTAAATTCTTGCAGAGTGTCAATGTTATCATTGATTATAGAAGACAGGGATAA
TACCTTTATCTCTGGCCACTCAAAAATGCAGTGCCAGGAGTGCTAAACCTAGAGGCCAATACTGATGACCTGGA
AGGTGATCCATATGATTGTCACCACAAAGTGCTTTTACACAAAAACTTGAAAATTTGAAAAACATGATTTTTTT
AAGTTCTCATCTCACCAGTCTTGGTGTTTATATTGCAAATCTATCAAAGTAGAAAATAATTTGTGCTGTATAC
AAATTACATGGGAACATAAAGGAGTGAGATCCTTCTGTGATAAAATGAATTCACGCACTCTGGTTACCCAACTA
CAGAACCTCCTTTGATCAGGCCAGTAGGTTGTGATGCAGGCTGGAGCCCCCGAATGCCCCACACACACTGCAGC
ATTGACCAGACCATCCGAAACCTGCGTCCCTGGTGATGTTCTCAAGCCTCGGAAGTGGCAAATGGAAATGATAT
GGCCGGTTGCGGTTGTAGGAGAGTTGTGACTTAGGCAGGAGTCGACCTCCTCAAGTAATGGAACGATTTCAAAG
GCAGGCTGCCCTGACCAAAAATATCTGCCATGAATAAAGGTGCCTGAAATCCTGCTAAAAAAAAAAAAAAAAAA
AAAAAA

The NOV7 8543.5 Dalton protein (SEQ ID NO:43) encoded by SEQ ID NO:42 is 76 amino acids in length and is presented using the one-letter code in Table 7B. The Psort profile for NOV7 predicts that this sequence has no known signal peptide and is likely to be localized in the cytoplasm with a certainty of 0.6500. In alternative embodiments, a NOV7 polypeptide is located to the mitochondrial matrix space with a certainty of 0.1000, or the lysosome (lumen) with a certainty of 0.1000.

Table 7B. NOV7 protein sequence (SEQ ID NO:43)

MAETDPKTVQDLTSVVQTLLQQMQDKFQTMSDQIIGRIDDMSSRIDDLEKNIADLMTQAGVEELESENKIPATQKS

The reverse complement for NOV7 is presented in Table 7C.

Table 7C. NOV7 reverse complement (SEQ ID NO:44)

TTTTTTTTTTTTTTTTTTTTTTAGCAGGATTTCAGGCACCTTTATTCATGGCAGATATTTTTGGTCAGGGCAGCCTG
CCTTTGAAATCGTTCCATTACTTGAGGAGGTCGACTCCTGCCTAAGTCACAACTCTCCTACAACCGCAACCGGCCATAT
CATTTCCATTTGCCACTTCCGAGGCTTGAGAACATCACCAGGGACGCAGGTTTCGGATGGTCTGGTCAATGCTGCAGTG
TGTGTGGGGCATTCGGGGGCTCCAGCCTGCATCACAACCTACTGGCCTGATCAAAGGAGGTTCTGTAGTTGGGTAACCA
GAGTGGTGAATTCATTTTATCACAGAAGGATCTCACTCCTTTATGTTCCCCATGTAATTTGTATACAGCACAAATTATT
TCTTACTTTGATAGATTTGCAATATAAACACCAAGACTGGTGAGATGAGAAACTTAAAAAATCATGTTTTCAAATTT
TCAAGTTTTTGTGTAAAAGCACTTTGTGGTGACAATCATATGGATCACCTTCCAGGTCATCAGTATTGGCCTCTAGGTT
TAGCACTCCTGGCACTGCATTTTTGAGTGGCCAGAGATAAAGGTATTATCCCTGTCTTCTATAATCAATGATAACATTG
ACACTCTGCAAGAATTTAGATAAGAAACAGTTTAAGTAAAATAGTGTTTTTTAAAAACTTGAGTCTACTACAAGTCGTT
GATCATATTCTAGCCAAGCAACTGGCATGGACTTAATATGGTCTCTTTTCCTTCTCTCTTTCTTTTCAAGTCACTAGCT
CAAAGGAAAGACTGATTGAGTTTTAAAATCCTGCCAGATTTTTTATGGTAACCATTGTAGATGAACCACGTAGGTAGGG
TCCTACTGAAGTATTAGAATCTCATGCAAAACTCTAACAAACGCCTCATTACAGAAGGCTATTCTGTTATTTTCTGTAC
TGTAAAAGTTCTGACACAAGACAGTGGCAGTGGTTACTTTTCATCGACTTTAGCATGTGATCTCAGGGACTCAGACATA
CGTCTAAGTTCTATTCTGAGTTTTGGCAACAGAGCAGTGACAGATATTTCTGAAATGAACAATTTTTAGGTGTTTCAGC
CATTTGAAAAGTATTGCCAACACACTATTTGGTGTTAGCTCAAAAGTCACGTTGTGCCAAGAATTAAAGAACTCTAAAG
TCTACAAACATCTTACTTCACCAAGACTAACTATAATTGAAGGGTTTACTATTTGTTTAATAAAAAATCACACATCAAC
TTTTATCCAAACAGCAACTACTACAAAAGGAATGACAACGAAAAAAATGACTTCACAGAAATACACTAAAAATCTGAC
AATGTTATGCAAGAACCGCCAAAGTTTTAGTGTTTAATAATGAATAGCACAACTGACCAAGATGTGAAGATA
CCATGTTCAAGAAACTGGGGGGAAATCACTCTACAAACTAACTATATACTAGGTGATAAGAATGCATACTTTATAATAT
AAAACCTGTCTACACATAGTAGTTAGCTGCAAAAAGCCATTCGATCTTCTCTTGGCTTGGAAAATGCCAGATTCCAGT
ATAAATTATTAGCAACCTTCAACTCTTTTGCGTGGCAGGTATCTTGTTTTCACTTTCCAGTTCTTCCACCCCAGCCTGT
GTCATGAGGTCCGCGATATTCTTTTCCAGATCATCAATGCGACTACTCATATCATCAATTCTCCCAATGATCTGGTCAG
ACATGGTCTGAAATTTATCTTGCATCTGCTGCAGGAGTGTCTGCACCACCGAGGTGAGGTCCTGCACGGTCTTGGGGTC
AGTCTCGGCCATCTCCCCGATGCCCAGCTTGGCGGTGATGTCTCAGACCGTAACCTACACTTCCGTTTGTCCGCTCAGT
CGCCCCGGGCCGCTGCTTCTCGCGACGAGACTGGCAGCTCGCGGGACCGCGGGGCATTATGGGAGTAGTAGTTTCTCCC
GCACCGGAAGGCCTCGAGGCCGCGCGCCC

BLASTP results for NOV7 are shown in Table 7D.

Table 7D. BLAST results for NOV7

Matching Entry (in SwissProt + SpTrEMBL)	Description	aa Length	% Identity	% Positive	E Value
HBP1_HUMAN; AF068754; AAC25186.1	HEAT SHOCK FACTOR BINDING PROTEIN 1. homo sapiens. 5/2000	76	76/76 (100%)	76/76, (100%)	4e-36
Q9CQZ1; AK018708; BAB31359.1	0610007A03RIK PROTEIN (SIMILAR TO HEAT SHOCK FACTOR BINDING PROTEIN1). mus musculus. 6/2001	76	67/76 (88%)	71/76, (93%)	8e-32
Q9VK90; AE003636; AAF53188.1	CG5446 PROTEIN. drosophila melanogaster. 5/2000	86	44/61 (72%)	51/61, (84%)	1e-18
Q9U3B7; Z77666; CAB01233.2	K08E7.2 PROTEIN. caenorhabditis elegans. 3/2001	80	36/54 (67%)	44/54, (81%)	3e-13
Q9FP22; AP003044; BAB19328.1	P0038C05.1 PROTEIN. oryza sativa. 3/2001	99	28/56 (50%)	42/56, (75%)	5e-10

A multiple sequence alignment is given in Table 7E, with the NOV7 protein of the invention being shown on lines 1 in a ClustalW analysis comparing NOV7 with related protein sequences of Table 7D.

Table 7E. Information for the ClustalW proteins:

1. SEQ ID NO:43, NOV7
2. SEQ ID NO:45, HBP1_HUMAN HEAT SHOCK FACTOR BINDING PROTEIN 1. 5/2000
3. SEQ ID NO:46, Q9CQZ1 0610007A03RIK PROTEIN mus musculus. 6/2001
4. SEQ ID NO:47, Q9VK90 CG5446 PROTEIN. drosophila melanogaster. 5/2000
5. SEQ ID NO:48, Q9U3B7 K08E7.2 PROTEIN. caenorhabditis elegans. 3/2001
6. SEQ ID NO:49, Q9FP22 P0038C05.1 PROTEIN. oryza sativa. 3/2001

```
NOV7          1  ------------------MAETPPKTVQDLTSVVQTLLQQWSLKFQTMSDQITQRIDDKS 42
HBP1_HUMAN    1  ------------------MAETPPKTVQDLTSVVQTLLQQWSDKFQTMSDQITQRIDDKS 42
Q9CQZ1        1  ------------------MAETPPKTMQDLTSLVQSLLQQWSKKFQTMSDQITQRIDDKS 42
Q9VK90        1  MTDLRNEMDSDLLDQNYSLNSNADPKNMQELTSYVQSLLQNWQDLNFQTMSDQITQRIDDVG 60
Q9U3B7        1  MSD-----EKSTTPTAQLDNPADQ-NMNDLTSLTQSYLQQWLDKFQTMSDQITQRIDDVT 54
Q9FP22        1  MAAPGSG-SGGIPIKADQDSDGSAQSTADMTAVVQNLLVQWQTRFQSVSDLISQIDDVG 59

NOV7         43  SRIDDLEKNIADLMTQAGVELESENKIPATQKS------ 76
HBP1_HUMAN   43  SRIDDLEKNIADLMTQAGVELESENKIPATQKS------ 76
Q9CQZ1       43  SRIDDLEKNIADLMTQAGVELDPENKIPTAQKS------ 76
Q9VK90       61  NRIDDLEKSIADLMNQAGHEGQGPEK-------------- 86
Q9U3B7       55  TRIDDLEKNINDLQSNQVDHPPSAQ-------------- 80
Q9FP22       60  ARIDELEQSINDLKVEMGTEGITPTKPKDEESKPAGSSAE 99
```

BLASTP domain results for NOV7 were collected from a proprietary database. The results are listed in Table 7F with the statistics and domain description.

Table 7F. Domain results for NOV7

ProDom Analysis	High Scores	Smallest Sum Probability P(N)
Sequences producing High-scoring Segment Pairs:		
prdm:42125 p36 (1) STE4_SCHPO // SEXUAL DIFFERENTIATION P...	78	0.0030
prdm:56790 p36 (1) BUD6_YEAST // BUD SITE SELECTION PROTE...	73	0.0059
prdm:53072 p36 (1) GAGY_DROME // RETROVIRUS-RELATED GAG P...	57	0.0074
prdm:35747 p36 (1) RLX2_SALTY // 22 KD RELAXATION PROTEIN...	69	0.017
prdm:8937 p36 (3) YOPE(3) // OUTER MEMBRANE VIRULENCE P...	64	0.073

```
prdm.42125 p36 (1) STE4_SCHPO//SEXUAL DIFFERENTIATION PROTEIN STE4. MEIOSIS, 264 aa.
Identities = 20/70 (28%), Positives = 42/70 (60%) for NOV7: 11-76, Sbjct: 62-131

>prdm.56790 p36 (1) BUD6_YEAST // BUD SITE SELECTION PROTEIN BUD6 (ACTIN INTERACTING
PROTEIN 3), 788 aa.
Identities = 12/50 (24%), Positives = 32/50 (64%) for NOV7: 20-69, Sbjct: 559-608
Identities = 7/24 (29%), Positives = 14/24 (58%) for NOV7: 3-26, Sbjct: 106-129

>prdm:53072 p36 (1) GAGY_DROME // RETROVIRUS-RELATED GAG POLYPROTEIN (TRANSPOSON
GYPSY). CORE PROTEIN; POLYPROTEIN; TRANSPOSABLE ELEMENT, 451 aa.
Identities = 12/38 (31%), Positives = 20/38 (52%) for NOV7: 5-41, Sbjct: 43-80
Identities = 8/19 (42%), Positives = 13/19 (68%) for NOV7: 58-76, Sbjct: 412-430

>prdm:35747 p36 (1) RLX2_SALTY // 22 KD RELAXATION PROTEIN  PLASMID, 194 aa.
Identities = 20/70 (28%), Positives = 37/70 (52%) for NOV7  7-74, Sbjct: 20-89

>prdm:8937 p36 (3) YOPE(3)//OUTER MEMBRANE VIRULENCE PROTEIN YOPE PLASMID, 219 aa.
Identities = 16/37 (43%), Positives = 22/37 (59%) for NOV7. 2-38, Sbjct: 111-147

PFAM HMM Domain Analysis

Scores for sequence family classification (score includes all domains):
Model    Description                                        Score    E-value  N
-------- -----------                                        -----    ------- ---
Leptin (InterPro)    Leptin                                  2.2         10   1

Parsed for domains:
Model    Domain  seq-f seq-t    hmm-f hmm-t      score  E-value
-------- ------- ----- -----    ----- -----      -----  -------
Leptin     1/1      20    42 ..      1    25 [.     2.2       10

PROSITE - Protein Domain Matches for Gene ID: NOV7
Pattern-ID: PKC_PHOSPHO_SITE PS00005 (Interpro) PDOC00005
Pattern-DE: Protein kinase C phosphorylation site
Pattern:    [ST].[RK]
NOV7 Position:   42-SSR; 73-TQK

Pattern-ID: CK2_PHOSPHO_SITE PS00006 (Interpro) PDOC00006
Pattern-DE: Casein kinase II phosphorylation site
Pattern:    [ST].[2}[DE]
NOV7 Position:   8-TVQD; 29-TMSD; 43-SRID

BLOCKS Analysis
AC#          Description                                        Strength  Score
BL01291A    0 NAD:arginine ADP-ribosyltransferases proteins       1609    1027
BL00058A    0 DNA mismatch repair proteins mutL / hexB / PM       1767    1001
BL00902A    0 Glutamate 5-kinase proteins.                        1549     994
BL01213C    0 Protozoan/cyanobacterial globins proteins.          1420     994
BL00579B    0 Ribosomal protein L29 proteins.                     1361     991
BL00487G    0 IMP dehydrogenase / GMP reductase proteins.         1525     989
BL00564F    0 Argininosuccinate synthase proteins.                1759     987
BL00154A    0 E1-E2 ATPases phosphorylation site proteins.        1268     983
```

The STE4_SCHPO //sexual differentiation protein STE4 is involved with meiosis. The bud6_yeast //bud site selection protein BUD6 (actin interacting protein 3) interacts with the cytoskeleton. The gagy_drome // retrovirus-related GAG polyperotein (transposon gypsy) is involved with viral core proteins; plyproteins; and transposable elements. Leptin is involved in fatty acid metabolism and body weight regulation.

Other BLAST results include sequences from the Patp database, which is a proprietary database that contains sequences published in patents and patent publications. Patp results include those listed in Table 7G.

Table 7G. Patp alignments of NOV7

Sequences producing High-scoring Segment Pairs:	% Identity	% Positive
patp:AAG19756 Arabidopsis thaliana protein fragment SEQ I...	54%	73%
patp:AAG19757 Arabidopsis thaliana protein fragment SEQ I...	60%	78%
patp:AAG19758 Arabidopsis thaliana protein fragment SEQ I..	60%	77%
patp:AAW60940 Streptococcus pneumoniae encoded polypeptid. .	32%	51%
patp:AAY43986 Mouse alcohol dehydrogenase #1 - Mus sp, 37. .	35%	57%
patp:AAY43987 Rat alcohol dehydrogenase #1 - Rattus sp, 3. .	35%	57%

NOV7 is expressed in at least the following tissues: Small intestine, skin, spleen, thyroid, placenta, colon, cervix, heart, uterus, tonsil, lung, parathyroid and others. This information was derived by determining the tissue sources of the sequences that were included in the invention including but not limited to SeqCalling sources, Public EST sources, Literature sources, and/or RACE sources. Based on the tissues in which NOV7 is most highly expressed, specific uses include developing products for the diagnosis or treatment of a variety of diseases and disorders. Additional disease indications and tissue expression for NOV7 is presented in Example 2.

The disclosed NOV7 nucleic acid encoding a novel protein includes the nucleic acid whose sequence is provided in Table 7A, or a fragment thereof. The invention also includes a mutant or variant nucleic acid any of whose bases may be changed from the corresponding base shown in Table 7A while still encoding a protein that maintains its activities and physiological functions, or a fragment of such a nucleic acid. The invention further includes nucleic acids whose sequences are complementary to those just described, including nucleic acid fragments that are complementary to any of the nucleic acids just described. The invention additionally includes nucleic acids or nucleic acid fragments, or complements thereto, whose structures include chemical modifications. Such modifications include, by way of nonlimiting example, modified bases, and nucleic acids whose sugar phosphate backbones are modified or derivatized. These modifications are carried out at least in part to enhance the chemical stability of the modified nucleic acid, such that they may be used, for example, as antisense binding nucleic acids in therapeutic applications in a subject. In the mutant or variant nucleic acids, and their complements, up to about 18 % percent of the bases may be so changed.

The disclosed NOV7 protein of the invention includes thenovel protein whose sequence is provided in Table 7B. The invention also includes a mutant or variant protein any of whose residues may be changed from the corresponding residue shown in Table 7B while still encoding a protein that maintains its activities and physiological functions, or a functional fragment thereof. In the mutant or variant protein, up to about 18 % percent of the residues may be so changed.

The invention further encompasses antibodies and antibody fragments, such as F_{ab} or $(F_{ab})_2$ that bind immunospecifically to any of the proteins of the invention.

The NOV7 nucleic acids and proteins of the invention are useful in potential therapeutic applications implicated in cancer including but not limited to breast cancer, ovarian cancer, and/or other pathologies and disorders. For example, a cDNA encoding the novel protein (NOV7) may be useful in cancer therapy, and thenovel protein (NOV7) may be useful when administered to a subject in need thereof. By way of nonlimiting example, the compositions of the present invention will have efficacy for treatment of patients suffering from cancer including but not limited to breast and ovarian cancer. The NOV7 nucleic acid encoding novel protein, of the invention, or fragments thereof, may further be useful in diagnostic applications, wherein the presence or amount of the nucleic acid or the protein are to be assessed.

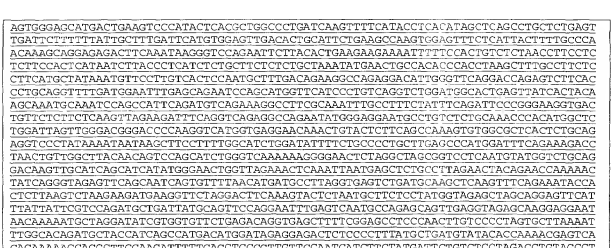
NOV7 nucleic acids and polypeptides are further useful in the generation of antibodies that bind immuno-specifically to the novel NOV7 substances for use in therapeutic or diagnostic methods. These antibodies may be generated according to methods known in the art, using prediction from hydrophobicity charts, as described in the "Anti-NOVX Antibodies" section below. The disclosed NOV7 protein has multiple hydrophilic regions, each of which can be used as an immunogen. In one embodiment, a contemplated NOV7 epitope is from about amino acids 1 to 10. In another embodiment, a NOV7 epitope is from about amino acids 20 to 25. In additional embodiments, NOV7 epitopes are from about amino acids 35 to 55, and from about amino acids 60 to 75. These novel proteins can be used in assay systems for functional analysis of various human disorders, which will help in understanding of pathology of the disease and development of new drug targets for various disorders.

NOV8

A disclosed NOV8 nucleic acid of 4204 nucleotides (also referred to as 24SC714) encoding a novel secreted protein is shown in Table 8A. An open reading frame was identified beginning with an ATG initiation codon at nucleotides 1911-1913 and ending with a TGA codon at nucleotides 2181-2183. A putative untranslated region upstream from the initiation codon and downstream from the termination codon is underlined in Table 8A, and the start and stop codons are in bold letters.

Table 8A. NOV8 nucleotide sequence (SEQ ID NO:50).

TTTTTGGAATATAAGTAGGGGTTTATTTGGGCCAGTCTTGAGGATTGAAACTTCAAAGCACAGATTAAAGTTATCCTGAAT
ATGTAGTCCGGTCCCACCAGCAACAGTTACAAATGGATTTTTAAAGGAAATAAAAGAAAAGGCAGTTCCTAAGTTGTTTAGC
AATAATTAACATATGAAAATAACATAAGCTATTGATCTGGCTATATGTTGTTCTTTGTTTCCTAAATTACAAGAAACGAAAG
ATAATGGGTGAGGCAGCTAGTTAGGAACTAAATGCTTTTAAACAATTCCCCCCACCCCCCACCGGTGTGGGTCCTGTGAGGG



AGTGGGAGCATGACTGAAGTCCCATACTCACGCTGGCCCTGATCAAGTTTTCATACCTCACATAGCTCAGCCTGCTCTGAGT
TGATTCTTTTTTATTGCTTTGATTCATGTGGAGTTGACACTGCATTCTGAAGCCAAGTGGAGTTTCTCATTACTTTTGCCCA
ACAAAGCAGGAAGAGACTTCAAATAAGGGTCCAGAATTCTTACACTGAAGAAGAAAATTTTTCCACTGTCTCTAACCTTCCTC
TCTTCCACTCATAATCTTACCCTCATCTCTGCTTCTCTCTGCTAAATATGAACTGCCACACACCGACTAAGCTTTGCCTCTC
CTTCAGCTCTATAAATTGTTCCTTGTCACTCCAATGCTTTGACAGAAGGCCAGAGAGACATTGGGTTCAGGACCAGAGTCCTTCAC
CCTGCAGGTTTTGATGGAATTTGAGCAGAATCCAGCATGGTCATCCTGTCAGGTCTGGATGGCACTGAGTTATCACTACA
AGCAAATGCAAATCCAGCCATTCAGATGTCAGAAAGGCCTTCGCAAATTTGCCTTTCTATTTCAGATTCCCGGGAAGGTGAC
TGTTCTCTTCTCAAGTAGAAGATTTCAGGTCAGAGGCCAGAATATGGGAGGAATGCCTGTCTCTGCAAACCCACATGGCTC
TGGATTAGTTGGGACGGGACCCCAAGGTCATGGTGAGGAACAAACTGTACTCTTCAGCCAAAGTGTGGCGCTCACTCTGCAG
AGGTCCCTATAAAATAATAAGCTTCCTTTGGCATCTGGATATTTTCTGCCCCTGCTTGAGCCCATGGATTTCAGAAAGACC
TAACTGTTGGCTTACAACAGTCCAGCATCTGGGTCAAAAAAGGGGAACTCTAGGCTAGCGGTCCTCAATGTATGGTCTGCAG
GACAAGTTGCATCAGCATCATATGGGAACTGGTTAGAAACTCAAATTAATGAGCTCTGCCTTAGAACTACAGAACCAAAAAC
TATCAGGGTAGAGTTCAGCAATCAGTGTTTTAACATGATGCCTTAGGTGAGTCTGATGCAAGCTCAAGTTTCAGAAATACCA
CTCTTAAGTCTAAGAAGATGAAGGTTCTAGGACTTCAAAGTACTCTAATGCTTCCTCTATGGTAGAGCTAGCAGGAGTTCAT
TTATTATTCGTCCAGATGCTGATTATGCAGTTCCAGGAATTTGAGTCAATGCCAGAGCAGTTGAGGTAGAGCAAGGAGGAAT
AACAAAAATGCTAGGATATCGTGGTGTTCTGAGACAGGTGAGCTTTTCGGAGCCTCCCAACTTGTCCCCTAGTGCTTAAAAT
TTGGCACAGATGCTACCATCAGCCATGACATGGATAGAGGAGACTCTCCCCTTTATGCTGATGTATACACCAAAACGAGTCCA
CAGAAAAAGCAGGCTTCCAAGATTTTCAGCTCCCGTTGTCCAATCATCTTCTATGATTCTGTCTCCTAGACCTGTAGCCT
TAAAGCAAGCTTATTTAAAATAATCTGCCAGCTCTGTTCAAAGAGATTTGTTCTCCTAAATTTGTCCAGACTGAAAACTG
CACAGTCCAAAGTTTAAGAGGTTATGTTAGGAGAAATTGAACATTATGTTTCCTACTGCTACTTAAATTTCCAGAGGCAT
TTACAAAAATTAAACATCAATGGGAAGCCAAGTCCTTTATGAAGCTAGCAATAGACATTGATCCTGTGATAATGTTATTATT
TTTCTTATTGCTCTTGTCAGTATGCATTTCATCATCGCTGGGTTGGATGAGTATAGGGCAGCATGGGAAAACAATGTTTATT
GACTTGCAGTTTCTAGGTGCTTTAAAAAAAGTTATGCACAGGTACATATGAGCATATTAAAGCTCTTAATTTGTGTTCTAA
TAATTTCTTCTTGAATCTCTAAAATTATGACACTACGATTAGCATTTTATTACCACATGTACAATCTATCCAGTCACCTTGA
AGTTAGATTAGATGGCATTCAAGTCACTCAGCACAGGTGAGTCAGACGGACTTTTGACCTCTTGTAAAATAGGAAAATAAA
GACAGTGACTTTATTTATAAGAAAAATGAACTTGGCCAACAACATTAGAGAATGCTTACTCATTCCTGTACCTAGACACAGAG
GAGCCTGGAACAGACCAGGAGAAATGAGACCATTATATACCCTATAATTACAACTTGTCTAATTGATCCAAGGGGAAGCAGA
GAAAGTTAACTGTAGGGCAGCAAGATGTAAACTTGGGAAGTCAGATAAGAATGGACCTGAAAGGGACCTTGAAAGGTATGC
AGGGGGCCTGGGCACAACTGCCAAGCATAATCAGACACTGTGTGAGAAGAGGAAGTAAGTCTAGTCCCAATCACTTAATAAG
TACAGATCTCCTAGGAAGAGGCTCTGGTACAGTATCCTTCCCCCGTCTTAAAGGGACATGGAGTCTCAGCCTCCCAGCAGGA
ATGTCTAGAGAAAAAGTATCTAGCTAATTTTGTGGGCAGGGTGAGGGAAGGAGAAATATTGTCTGGCTTAGTAGAGAGTGTG
GTCTCCACAGTAACACAGATCCTGATGTGACATTTGAGGCAGCATCCTTCCTGTGGTCAAGACTTTTCCTCCTCGTGCATT
CTGGATCCCTTCCCTGGTGTCTTTTCAGGGCATCAATTACCCCATCTCTCTCTTATCTAGTCAACCCTTTCCTCGCAATCTT
CCCCAAAACACTTAAACAGCTCAAGCTTTCCCCACCTTAAAAATATCTTCCCTCTACCCCACACTTCCTGCAGCTACAGCA
CTCTCTCCTCCTCCTCACACCCAAAGTTTTCCAGAAAATTATCCATCCTTGCCATCTCCATATGCTCCCCTCCCACTCCTCA
ATTCACCTCGCTCTGTCTTCCACTCCTGTCACAGGCTTTAAAAAGCCACTGCAATCATTAGGTGACCTGTCTATTGCCAAAG
TCTCAGGACATTTTCAATTCTACCTTACTTGAAACCTCCGCAGTGTGAAGGTCACTCCTTCCATCTATGCTCCTTCCTGGGT
TCTTGGGGCTCCACAATCCTCGTGGCTTCCTCCTACCCACCTGCCTGCTTATTCATTTATTCTGCAGGCTCCTTCCCCTAC
CCGACATGCCAGAGTTCCTACAAGCTTCAGGAGTCGTCCTTGACTTCTCCCTCTTCCTCACCACTCTCCAATCCAAAACATC
ACCAAATCTTGTTAATTTGGGTCCTTTGGTATTTGTTTATTCTGTCGGTTTTTTTCTGTCTTCACTCCTCTCATTCTCTAAG
AGCTGCTATAGCCTCCTTCACAACAAAGAGAGAGAGCTGCCTAAAGTCACCCAGCTAATGAATGATGACTAGGAGTGGTTCC
CAGATATTTATCCCTTACTGCTGTGGAGGTCCTCATCACCCTAATAGAATCACTCTTTATTCACAAAAGTAGAAAATTAA
TTTTGGATACATCATTTATTATCAAGATGTTGTTGAGGAAAAATAGGGTCATGTAAGGTCCCTCTCAGCATTTCCTTCAAG
TTGCAAGAATTAGAAAAACAGAGACAAGATTCTATGTGTGTCCTCAGAAGACCTTCCTGAGGACCATTCCCCTAGGAACTTA
AAAAAATTAAGCCTCCAACTCTTCCATCTTAACTGTGTAACAGAGGAAGGTGATGACAAGAGGAAGGAGACAAGCAAGAGT
CAGACTTCGAAGGCTTGGCAGCCACTGTCAGCAAGAGGTGAGAACAGCAGACAAGACAGCAACACACTCCTGAAATAATCAATC
CATACGGACTGCCATGTGAAATGTGGAGCAGACTAGTTCTAAATGGCTCCAGGAGGCAAAATAAGACTCAAGAGAAGTTACT
GGTAGATTTCAACCCAATGTGA

The NOV8 nucleic acid was identified on chromosome 3 by comparing a NOV8 nucleic acid to the human genome. Exons were predicted by homology and the intron/exon boundaries were determined using standard genetic rules. Exons were further selected and refined by means of similarity determination using multiple BLAST (for example, tBlastN, BlastX, and BlastN) searches, and, in some instances, GeneScan and Grail. Expressed sequences from both public and proprietary databases were also added when available to further define and complete the gene sequence. The DNA sequence was then manually corrected for apparent inconsistencies thereby obtaining the sequences encoding the full-length protein. The NOV8 nucleic acid was further localized to the 3p22 region, a locus associated with cancer, *e.g.* esophageal (OMIM 604050), hepatoblastoma (OMIM 116806), lung (OMIM 604050), and ovarian carcinoma

(OMIM 116806), and psuedo-Zellweger syndrome (OMIM 604054). NOV8 is useful as a marker for these diseases.

A disclosed NOV8 polypeptide (SEQ ID NO:51) encoded by SEQ ID NO:50 has 90 amino acid residues and is presented in Table 8B using the one-letter amino acid code. SignalP, Psort and/or Hydropathy results predict that NOV8 has a signal peptide and is likely to be secreted with a certainty of 0.8200. The most likely cleavage site for a NOV8 peptide is between amino acids 61 and 62, at SLG-WM. NOV8 has a molecular weight of 10,474.6 Daltons.

Table 8B. Encoded NOV8 protein sequence (SEQ ID NO:51).

```
MLGEIEHYVFLLLLKFPFAFTKIKHQWEAKSFMKLAIDIDPVIMLLFFLLLLSVCISSSLGWMSIGQHGKTMFIDLQFLGAL
KKVMMRYI
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The presence of identifiable domains in NOV8, as well as all other NOVX proteins, was determined by searches using software algorithms such as PROSITE. DOMAIN, Blocks, Pfam, ProDomain, and Prints, and then determining the Interpro number by crossing the domain match (or numbers) using the Interpro website (http:www.ebi.ac.uk/ interpro). DOMAIN results for NOV8 as disclosed in Tables 1E, were collected from the Conserved Domain Database (CDD) with Reverse Position Specific BLAST analyses. This BLAST analysis software samples domains found in the Smart and Pfam collections.

Prodom domain analysis of the NOV8 polypeptide indicates that the NOV8 polypeptide has 11 of 23 (47%) identical to, and 14 of 23 (60%) positive with, the 40 aa p36 (12) ATCD(5) ATCE(4) ATCB(2) – calcium reticulum calcium-transporting ATPase type hydrolase transport transmembrane endoplasmic class (prdm:2196, Expect = 0.36); 28 of 84 (33%) identical to, and 38 of 84 (45%) positive with, the 1769 aa p36 (1) YJK9_YEAST - hypothetical 200.0 kD protein in GZF3-SME1 intergenic region, hypothetical protein (prdm:57835, Expect = 0.36); 11 of 32 (34%) identical to, and 18 of 32 (56%) positive with, the 68 aa p36 (2) G49(1) G49B(1) – glycoprotein mast cell surface precursor signal transmembrane immunoglobulin fold GP49A (prdm:15250, Expect = 0.58); 9 of 23 (39%) identical to, and 17 of 23 (73%) positive with, the 41 aa p36 (1) WNT1_CAEEL - WNT-1 protein precursor (prdm:47898, Expect = 0.58); and 15 of 46 (32%) identical to, and 26 of 46 (56%) positive with, the 89 aa p36 (1) SAPB_HAEIN – peptide transport system permease protein SAPB (prdm:35160, Expect = 1.1). Table 8C lists the domain description from DOMAIN analysis results against NOV8. This indicates that the NOV8 sequence has properties similar to those of other proteins known to contain this domain.

Table 8C. Domain Analysis of NOV8

```
ProDom Protein Domain Analysis                                    High   Smallest Sum Probability
Sequences producing High-scoring Segment Pairs:                   Score  P(N)
prdm:2196  p36 (12) ATCD(5) ATCE(4) ATCB(2)  - CALCIUM R...      52     0.30
prdm:57835 p36 (1) YJK9_YEAST - HYPOTHETICAL 200.0 KD PR...      68     0.30
prdm:15250 p36 (2) G49(1) G49B(1)  - GLYCOPROTEIN MAST ...       50     0 44
prdm:47898 p36 (1) WNT1_CAEEL - WNT-1 PROTEIN PRECURSOR....     50     0.44
prdm:35160 p36 (1) SAPB_HAEIN - PEPTIDE TRANSPORT SYSTEM...     55     0.66

BLOCKS Protein Domain Analysis
AC#          Description                                          Strength  Score
BL00456D    0 Sodium:solute symporter family proteins.            1174    1038
BL01271B    0 Sodium:sulfate symporter family proteins.           1480    1033
BL00790A    0 Receptor tyrosine kinase class V proteins.          1390    1031
BL00284A    0 Serpins proteins.                                   1308    1029
BL01313A    0 Lipoate-protein ligase B proteins.                  1390    1018

PROSITE - Protein Domain Analysis
Protein Domain Matches for Gene ID: NOV08
No PROSITE patterns found
```

In a search of public sequence databases, the NOV8 amino acid sequence had no hits with the Expect value set at 1.0. Public amino acid databases include the GenBank databases, SwissProt, PDB and PIR.

Other BLAST results include sequences from the Patp database, which is a proprietary database that contains sequences published in patents and patent publications. BLASTP analysis again the NOV8 protein shows that the NOV8 protein has 18 of 28 aa residues (64%) identical to, and 18 of 28 aa residues (64%) positive with, the 78 aa Zea mays protein fragment SEQ ID NO: 30302 of patent EP1033405-A2 (patp: AAG26008, Expect = 0.097);14 of 30 aa residues (46%) identical to, and 16 of 30 aa residues (53%) positive with, the 51 aa Human secreted protein sequence encoded by gene 65 SEQ ID NO:188 (patp:AAY91515, Expect = 0.50); 14 of 30 aa residues (46%) identical to, and 16 of 30 aa residues (53%) positive with, the 50 aa Human secreted protein sequence encoded by gene 65 SEQ ID NO:329 (patp:AAY91656, Expect = 0.50); 21 of 64 aa residues (32%) identical to, and 32 of 64 aa residues (50%) positive with, the 997 aa Human shear stress-response protein SEQ ID NO: 28 (patp:AAB90764, Expect = 0.91); 13 of 31 aa residues (41%) identical to, and 19 of 31 aa residues (61%) positive with, the 52 aa Gene 9 human secreted protein homologous amino acid sequence #123 - Chlorella vulgaris (patp:AAB34919, Expect = 1.0); and 14 of 43 aa residues (32%) identical to, and 22 of 43 aa residues (51%) positive with, the 46 aa Human secreted protein sequence encoded by gene 4 SEQ ID NO:64 (patp:AAB34580, Expect = 2.7). Patp results include those listed in Table 8D.

Table 8D. Patp alignments of NOV8

Sequences producing High-scoring Segment Pairs:	High Score	Smallest Sum Prob. P(N)
patp:AAY91515 Human secreted protein sequence encoded by ...	59	0.39
patp:AAY91656 Human secreted protein sequence encoded by ...	59	0.39
patp:AAB90764 Human shear stress-response protein SEQ ID ...	70	0.60
patp:AAB34919 Gene 9 human secreted protein homologous am...	56	0.64
patp:AAB34580 Human secreted protein sequence encoded by ...	52	0.93

The NOV8 protein domain information and chromosomal mapping suggest that NOV8 is a cancer-associated secreted protein. As such, it is useful as a diagnostic tool for the onset and or progression of cancer, such as esophageal, hepatoblastoma, lung, and ovarian carcinoma.

The disclosed NOV8 nucleic acid encoding a secreted protein includes the nucleic acid whose sequence is provided in Table 8A, or a fragment thereof. The invention also includes a mutant or variant nucleic acid any of whose bases may be changed from the corresponding base shown in Table 8A while still encoding a protein that maintains its secreted protein-like activities and physiological functions, or a fragment of such a nucleic acid. The invention further includes nucleic acids whose sequences are complementary to those just described, including nucleic acid fragments that are complementary to any of the nucleic acids just described. The invention additionally includes nucleic acids or nucleic acid fragments, or complements thereto, whose structures include chemical modifications. Such modifications include, by way of nonlimiting example, modified bases, and nucleic acids whose sugar phosphate backbones are modified or derivatized. These modifications are carried out at least in part to enhance the chemical stability of the modified nucleic acid, such that they may be used, for example, as antisense binding nucleic acids in therapeutic applications in a subject.

The disclosed NOV8 protein of the invention includes the secreted protein -like protein whose sequence is provided in Table 8B. The invention also includes a mutant or variant protein any of whose residues may be changed from the corresponding residue shown in Table 8B while still encoding a protein that maintains its secreted protein -like activities and physiological functions, or a functional fragment thereof.

The invention further encompasses antibodies and antibody fragments, such as F_{ab} or $(F_{ab})_2$, that bind immunospecifically to any of the proteins of the invention.

The above defined information for this invention suggests that this secreted protein -like protein (NOV8) may function as a member of a secreted protein family. Therefore, the NOV8 nucleic acids and proteins identified here may be useful in potential therapeutic applications implicated in (but not limited to) various pathologies and disorders as indicated below. The potential therapeutic applications for this invention include, but are not limited to: cancer

research tools, for all tissues and cell types composing (but not limited to) those defined here, including esophagus, liver, lung and ovary.

The NOV8 nucleic acids and proteins of the invention are useful in potential therapeutic applications implicated in cancer including but not limited to esophageal, liver, lung and ovary and/or other pathologies and disorders. For example, a cDNA encoding the secreted protein-like protein (NOV8) may be useful in cancer therapy, and the secreted protein -like protein (NOV8) may be useful when administered to a subject in need thereof. By way of nonlimiting example, the compositions of the present invention will have efficacy for treatment of patients suffering from cancer including but not limited to esophageal, hepatic, lung and ovarian cancer. The NOV8 nucleic acid encoding secreted protein -like protein, and the secreted protein -like protein of the invention, or fragments thereof, may further be useful in diagnostic applications, wherein the presence or amount of the nucleic acid or the protein are to be assessed.

NOV8 nucleic acids and polypeptides are further useful in the generation of antibodies that bind immuno-specifically to the novel NOV8 substances for use in therapeutic or diagnostic methods. These antibodies may be generated according to methods known in the art, using prediction from hydrophobicity charts. as described in the "Anti-NOVX Antibodies" section below. The disclosed NOV8 protein has multiple hydrophilic regions, each of which can be used as an immunogen. In one embodiment, a contemplated NOV8 epitope is from about amino acids 1 to 30 In another embodiment, a NOV8 epitope is from about amino acids 18 to 35. In additional embodiments, NOV8 epitopes are from about amino acids 65 to 90. These novel proteins can be used in assay systems for functional analysis of various human disorders, which will help in understanding of pathology of the disease and development of new drug targets for various disorders.

NOV9

A disclosed NOV9 nucleic acid of 3111 nucleotides (also referred to as 6CS060) encoding a novel Kelch-like protein is shown in Table 9A. An open reading frame was identified beginning with an ATG initiation codon at nucleotides 1-3 and ending with a TAA codon at nucleotides 1708-1710. A putative untranslated region downstream from the termination codon is underlined in Table 9A, and the start and stop codons are in bold letters.

Table 9A. NOV9 nucleotide sequence (SEQ ID NO:52).

ATGAATGCCACCAGATCTGAAGAGCAGTTCCATGTTATAAACCACGCAGAGCAAACTCTTCGTAAAATGGAGAACTACTTG
AAAGAGAAACAACTATGTGATGTGCTACTGATTGCAGGACACCTCCGCATCCCAGCCCATAGGTTGGTTCTCAGCGCAGTG
TCTGATTATTTTGCTGCAATGTTTACTAATGATGTGCTTGAAGCCAAACAAGAAGAGGTCAGGATGGAAGGAGTAGATCCA
AATGCACTAAATTCCTTGGTGCAGTATGCTTACACAGGAGTCCTGCAATTGAAAGAAGATACCATTGAAAGTTTGCTGGCT

GCAGCTTGTCTTCTGCAGCTGACTCAGGTCATTGATGTTTGCTCCAATTTTCTCATAAAGCAGCTCCATCCTTCAAACTGC
TTAGGGATTCGATCATTTGGAGATGCCCAAGGCTGTACAGAACTTCTGAACGTGGCACACAAATACACTATGGAACACTTC
ATTGAGGTAATAAAAAACCAAGAATTCCTCCTGCTTCCAGCTAATGAAATTTCAAAACTTCTGTGCAGTGATGACATTAAT
GTGCCTGATGAAGAGACCATTTTTCATGCTCTAATGCAGTGGGTGGGGCATGATGTGCAGAATAGGCAAGGAGAACTGGGG
ATGCTGCTTTCTTACATCAGACTGCCATTACTCCCACCACAGTTACTGGCAGATCTTGAAACCAGTTCCATGTTTACTGGT
GATCTTGAGTGTCAGAAGCTCCTGATGGAAGCTATGAAGTATCATCTTTTGCCTGAGAGAAGATCCATGATGCAAAGCCCT
CGGACAAAGCCTAGAAAATCAACTGTGGGGGCACTTTATGCTGTAGGAGGCATGGATGCTATGAAAGGTACTACTACTATT
GAAAAATATGACCTCAGGACCAACAGTTGGCTACATATTGGCACCATGAATGGCCGTAGGCTTCAATTTGGAGTCGCAGTT
ATTGATAATAAGCTCTATGTCGTGGGAGGAAGAGACGGTTTAAAAACTTTGAATACAGTGGAATGTTTTAATCCAGTTGGC
AAAATCTGGACTGTGATGCCTCCCATGTCAACACATCGGCACGGCTTAGGTGTAGCCACTCTTGAAGGACCAATGTATGCT
GTAGGTGGTCATGATGGATGGAGCTATCTAAATACTGTAGAAAGATGGGACCCTGAGGGACGACAGTGGAATTACGTAGCC
AGTATGTCAACTCCTAGAAGCACAGTTGGTGTTGTTGCATTAAACAACAAATTATATGCTATTGGTGGACGTGATGGAAGT
TCCTGCCTCAAATCAATGGAATACTTTGACCCACACACTAACAAGTGGAGTTTGTGTGCTCCAATGTCCAAAAGACGTGGA
GGTGTGGGAGTTGCCACATACAATGGATTCTTATATGTTGTAGGGGGGCATGATGCCCCTGCTTCCAACCATTGCTCCAGG
CTTTCTGACTGTGTGGAACGGTATGATCCAAAAGGTGATTCATGGTCAACTGTGGCACCTCTGAGTGTTCCTCGAGATGCT
GTTGCTGTGTGCCCTCTTGGAGACAAACTCTACGTGGTTGGAGGATATGACGGACATACTTATTGAACACAGTTGAGTCA
TATGATGCACAGAGAAATGAATGGAAGAGGAAGTTCCTGTTAACATTGGAAGAGCTGGTGCATGTGTTGTAGTGGTGAAG
CTACCCTAAAGCTATCTATCTTTATCAAATGGAATGAAACTAGATAATTTCAAGAAACTGAGTAGGACAAAGGGAGAAAGA
AATACATGTTCTTTTCCTGCAATTAATAATCAGACTGGAAAATTGTTGTATCATTTTAATTTGTAGTTACAATTGCTTTC
ATTCGTGAAGCCGAAACGTTTTTAAACATGAATTACATATGAATTATTAAGCATATGTGCTTTCGCAGCTGATAATATAAA
AGGAAATCCCACAGTCTAGATATAGCCCCATTACTACAAAATGCTAAAATATTTAATGAAAATTGATGGTGGCCACAGTGT
GCAGGTTATAAAAGCATTAATACATTTCAAGGTAAGAGCCTTAAAAGTTAAAAACATTTTCAGTTTTTTTTTAAAAAACGT
ACTCTTATTATCTGGAACATAGAAATATAAAAGGTAACATCTAAAGCTTTAGAATAGTGTAATTTTAGTAAGCCATTATTC
TCCTATTCAAATAATATCCCAAAGAGCTAAACAATTCCTTACATTTACCAAGAGGAAAGCTTTTACTGTGTTGAAGCTAAA
AAAATAATGGCTCTTTGACAAAACTTGTTATGTTGATCGCGGTATGTCAAAATTTTTACAGGTTTGCTCATCTGCCAGAGC
ACACATATAAATTTGGTATTTCTTAACATATCATCTTGTTAGATTTGTTACCAGTAAAATATTACTGTAATTTCATATACA
CAGTCTATACAATGAAATAATGAATATTTATCATATTGATACAAACTGTGACCTCAGCTTCAGAGTGTCAGGGCCTCACTT
GTATAGAATGTAATGTTCTCCTCAAACATTTATGTTAACTCTATAAACAAATATCGTTAAGTTAAACAAGTTTTCAAAAAC
AAAACAATTTTTAAAGTACCTTAAAATTGAGGATGTTACTCAGTGTTAACACATGGGAACACCAAAATATTCAATAAGCCT
GGTCAATTCTATAGTTATCTTTTTTGTACCAACACATGCTTTTCTGTTACTGTTATATTATCCAGTAGAAAATGTTAGGAT
ATGTGTGCTATATAAAAAAAAAAAAAGACTTGTTAAGTTTTAAAATAACAAAAATGGCTAGTTGAATAGTATTTTATGTGT
AATTCTTCCATTATTCTGTTTAATTATACAACTAAGATGAAAATTGAAAAACCCTTTGTGAAAGTAACTTTTCAAGTAA
ATGCACAACTTTAGAATTTCTACAAATAAGTTCTTTTAAACAGTCTTTTTATTGTGGATTGTGAAATCAAAATCTGGAGAA
ATGCTTATAAAATATACTACTAGCTTTAAGTTTTAAGAAAGAAGAACGTAAGTTGTACAAAGATATTTGTACTTTGACAA
ACTGAATTTAAATAAACTTTATTTCCTCTCAAA

The NOV9 nucleic acid was identified on the human X chromosome by comparing the NOV9 nucleic acid to the human genome. Exons were predicted by homology and the intron/exon boundaries were determined using standard genetic rules. Exons were further selected and refined by means of similarity determination using multiple BLAST (for example, tBlastN, BlastX, and BlastN) searches, and, in some instances, GeneScan and Grail. Expressed sequences from both public and proprietary databases were also added when available to further define and complete the gene sequence. The DNA sequence was then manually corrected for apparent inconsistencies thereby obtaining the sequences encoding the full-length protein. The NOV9 nucleic acid was further mapped to the q13 region of the X chromosome. This locus is associated with Menkes disease (OMIM 300011), myoglobinuria/hemolysis due to PGK deficiency (OMIM 311800), Wieacker-Wolff syndrome (OMIM 314580) and/or other diseases/disorders. NOV9 is a useful marker for these and/or other diseases/disorders.

In a search of public sequence databases, the NOV9 nucleic acid sequence has 2751 of 2767 bases (99 %) identical to a human Kelch-4 cDN (Accession No. XM039746). Public nucleotide databases include all GenBank databases and the GeneSeq patent database.

A disclosed NOV9 polypeptide (SEQ ID NO:53) encoded by SEQ ID NO:52 has 569 amino acid residues and is presented in Table 9B using the one-letter amino acid code. SignalP, Psort and/or Hydropathy results predict that NOV9 does not contain a known signal peptide and is likely to be localized endoplasmic reticulum (membrane with a certainty of 0.6000. In alternative embodiments, the NOV9 protein is localized to a microbody (peroxisome) with a certainty of 0.3000; the mitochondrial inner membrane with a certainty of 0.1000; or the plasma membrane with a certainty of 0.1000. NOV9 has a molecular weight of 63292.0 Daltons.

Table 9B. Encoded NOV9 protein sequence (SEQ ID NO:53).

```
MNATRSEEQFHVINHAEQTLRKMENYLKEKQLCDVLLIAGHLRIPAHRLVLSAVSDYFAAMFTNDVLEAKQEEVRMEGVDP
NALNSLVQYAYTGVLQLKEDTIESLLAAACLLQLTQVIDVCSNFLIKQLHPSNCLGIRSFGDAQGCTELLNVAHKYTMEHF
IEVIKNQEFLLLPANEISKLLCSDDINVPDEETIFHALMQWVGHDVQNRQGELGMLLSYIRLPLLPPQLLADLETSSMFTG
DLECQKLLMEAMKYHLLPERRSMMQSPRTKPRKSTVGALYAVGGMDAMKGTTTIEKYDLRTNSWLHIGTMNGRRLQFGVAV
IDNKLYVVGGRDGLKTLNTVECFNPVGKIWTVMPPMSTHRHGLGVATLEGPMYAVGGHDGWSYLNTVERWDPEGRQWNYVA
SMSTPRSTVGVVALNNKLYAIGGRDGSSCLKSMEYFDPHTNKWSLCAPMSKRRGGVGVATYNGFLYVVGGHDAPASNHCSR
LSDCVERYDPKGDSWSTVAPLSVPRDAVAVCPLGDKLYVVGGYDGHTYLNTVESYDAQRNEWKEEVPVNIGRAGACVVVVK
LP
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The reverse complement for NOV9 is presented in Table 9C.

Table 9C. NOV9 reverse complement (SEQ ID NO:54)

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TTTGAGAGGAAATAAAGTTTATTTAAATTCAGTTTGTCAAAGTACAAATATCTTTGTACAACTTACGTTCTTCTTTCTTAA
AACTTAAAAGCTAGTAGTATATTTTATAAGCATTTCTCCAGATTTTGATTTCACAATCCACAATAAAAAGACTGTTTAAAA
GAACTTATTTGTAGAAATTCTAAAGTTGTGCATTTACTTGAAAAGTTACTTTCACAAAGGGTTTTTCAAATATTTCATCTTA
GTTGTATAATTAAACAGAATAAATGGAAGAATTACACATAAAATACTATTCAACTAGCCATTTTTGTTATTTAAAACTTA
ACAAGTCTTTCTTTTTTTTTAATATAGCACACATATCCTAACATTTTCTACTGGATAATATAACAGTAACAGAAAAGCATGT
GTTGGTACAAAAAAGATAACTATAGAATTGACCAGGCTTATTGAATATTTTGGTGTTCCCATGTGTTAACACTGAGTAACA
TCCTCAATTTTAAGGTACTTTAAAAAATTGTTTTGTTTTTGAAAACTTGTTTAACTTAACGATATTTGTTTATAGAGTTAAC
ATAAATGTTTGAGGAGAACATTACATTCTATACAAGTGAGGCCCTGACACTCTGAAGCTGAGGTCACAGTTTGTATCAATA
TGATAAATATTCATTATTTCATTGTATAGACTGTGTATATGAAATTACAGTAATATTTTACTGGTAACAAATCTAACAAGA
TAATATGTTAAGAAATACCAAATTTATATGTGTGCTCTGGCAGATGAGCAAACCTGTAAAAATTTGACATACCGCGATCA
ACATAACAAGTTTTGTCAAAGAGCCATTATTTTTTTAGCTTCAACACAGTAAAAGCTTTCCTCTTGGTAAATGTAAGGAAT
TGTTTAGCTCTTTGGGATATTATTTGAATAGGAGAATAATGGCTTACTAAAAATCACACTATTCTAAGCTTTAGATGTTAC
CTTTTATATTTCTATGTTCCAGATAATAAGAGTACGTTTTTTAAAAAAAAACTGAAAATGTTTTTAACTTTTAAGGCTCTT
ACCTTGAAATGTATTAATGCTTTTATAACCTGCACACTGTGGCCACCATCAATTTTCATTAAATATTTTAGCATTTTGTAG
TAATGGGGCTATATCTAGACTGTGGGATTTCCTTTTATATTATCAGCTGCGAAAGCACATATGCTTAATAATTCATATGTTA
ATTCATGTTTAAAAACGTTTCGGCTTCACGAATGAAAGCAATTGTAACTACAAATTAAAATGATACAACAATTTTCCGCTC
TGATTATTAATTGCAGGAAAAAGAACATGTATTTCTTTCTCCCTTTTGTCCTACTCAGTTTCTTTGAAAATTATCTAGTTTCAT
TCCATTTGATAAAGATAGATAGCTTTAGGGTAGCTTCACACTCACAACACATGCACCAGCTCTTCCAATGTTAACAGGAAC
TTCCTCTTTCCATTCATTTCTCTGTGCATCATATGACTCAACTGTGTTCAAATAAGTATGTCCGTCATATCCTCCAACCAC
GTAGAGTTTGTCTCCAAGAGGGCACACAGCAACAGCATCTCGAGGAACACTCAGAGGTGCCACAGTTGACCATGAATCACC
TTTTGGATCATACCGTTCCACACAGTCAGAAAGCCTGGAGCAATGGTTGGAAGCAGGGCATCATGCCCCCTACAACATA
TAAGAATCCATTGTATGTGGCAACTCCCACACCTCCACGTCTTTTGGACATTGGAGCACACAAACTCCACTTGTTAGTGTG
TGGGTCAAAGTATTCCATTGATTTGAGGCAGGAACTTCCATCACGTCCACCAATAGCATAATTGTTGTTGATTTGAATGCAAA
AACACCAACTGTGCTTCTAGGAGTTGACATACTGGCTACGTAATTCCACTGTCGTCCCTCAGGGTCCCATCTTTCTACAGT
ATTTAGATAGCTCCATCCATCATGACCACCTACAGCATACATTGGTCCTTCAAGAGTGGCTACACCTAAGCCGTGCCGATG
TGTTGACATGGGAGGCATCACAGTCCAGATTTTGCCAACTGGATTAAAACATTCCACTGTATTCAAAGTTTTTAAACCGTC
TCTTCCTCCCACGACATAGAGCTTATTATCAATAACTGCGACTCCAAATTGAAGCCTACGGCCATTCATGGTGCCAATATG
TAGCCAACTGTTGGTCCTGAGGTCATATTTTTCAATAGTAGTAGTACCTTTCATAGCATCCATGCCTCCTACAGCATAAAG
TGCCCCCACAGTTGATTTTCTAGGCTTTGTCCGAGGGCTTTGCATCATGGATCTTCTCTCAGGCAAAAGATGATACTTCAT
AGCTTCCATCAGGAGCTTCTGACACTCAAGATCACCAGTAAACATGGAACTGTTTCAAGATCTGCCAGTAACTGTGGTGG
GAGTAATGGCAGTCTGATGTAAGAAAGCAGCATCCCAGTTCTCCTTGCCTATTCTGCACATCATGCCCCACCCACTGCAT
TAGAGCATGAAAAATGGTCTCTTCATCAGGCACATTAATGTCATCACTGCACAGAAGTTTTGAAATTTCATTAGCTGGAAG
CAGGAGGAATTCTTGGTTTTTTATTACCTCAATGAAGTGTTCCATAGTGTATTTGTGTGCCACGTTCAGAAGTTCTGTACA
GCCTTGGGCATCTCCAAATGATCGAATCCCTAAGCAGTTTGAAGGATGGAGCTGCTTTATGAGAAAATTGGAGCAAACATC
```

```
AATGACCTGAGTCAGCTGCAGAAGACAAGCTGCAGCCAGCAAACTTTCAATGGTATCTTCTTTCAATTGCAGGACTCCTGT
GTAAGCATACTGCACCAAGGAATTTAGTGCATTTGGATCTACTCCTTCCATCCTGACCTCTTCTTGTTTGGCTTCAAGCAC
ATCATTAGTAAACATTGCAGCAAAATAATCAGACACTGCGCTGAGAACCAACCTATGGGCTGGGATGCGGAGGTGTCCTGC
AATCAGTAGCACATCACATAGTTGTTTCTCTTTCAAGTAGTTCTCCATTTTACGAAGAGTTTGCTCTGCGTGGTTTATAAC
ATGGAACTGCTCTTCAGATCTGGTGGCATTCAT
```

In a search of public sequence databases, the NOV9 amino acid sequence has 431 of 569 amino acid residues (76%) identical to, and 500 of 569 residues (88 %) positive with, the 569 amino acid residue human Kelch-like protein-1. Public amino acid databases include the GenBank databases, SwissProt, PDB and PIR.

It was also found that NOV9 had homology to the amino acid sequences shown in the BLASTP data listed in Table 9D.

Table 9D. BLAST results for NOV9

Gene Index/ Identifier	Protein/ Organism	Length (aa)	Identity (%)	Positives (%)	Expect
Q9C0H6; AB051474; BAB21778.1	KIAA1687 PROTEIN (FRAGMENT). homo sapiens. 6/2001	728	569/569 (100%)	569/569, (100%)	0.0
Q9Y3J5; AL035424; CAB39994.1	DA22D12.1. homo sapiens. 6/2001	569	569/569 (100%)	569/569, (100%)	0.0
KHL1_HUMAN; AF252283; AAF81719.1	KELCH-LIKE PROTEIN 1. homo sapiens. 10/2000	748	431/569 (76%)	500/569, (88%)	0.0
KHL1_MOUSE; AF252281; AAF81717.1	KELCH-LIKE PROTEIN 1. mus musculus. 10/2000	751	430/569 (76%)	497/569, (87%)	0.0
Q9H955; AK023057; BAB14382.1	CDNA FLJ12995 FIS, CLONE NT2RP3000233, weakly similar to ring canal protein. homo sapiens. 6/2001	411	411/411 (100%)	411/411, (100%)	0.0

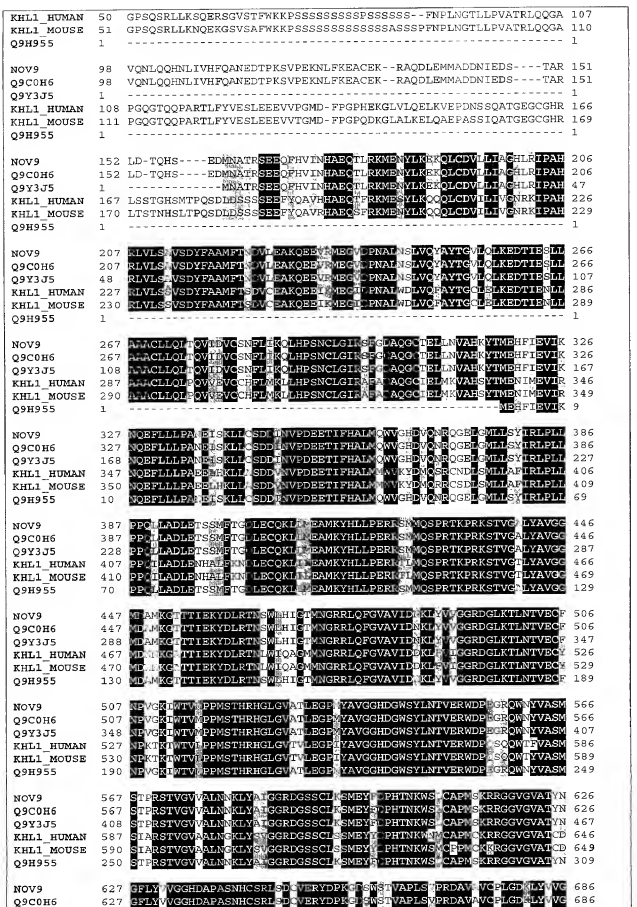
A multiple sequence alignment is given in Table 9E, with the NOV9 protein of the invention being shown on lines 1 in a ClustalW analysis comparing NOV9 with related protein sequences of Table 9D.

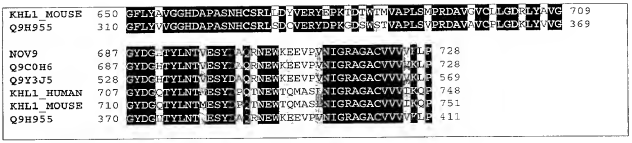
Table 9E. Information for the ClustalW proteins:

```
1.  SEQ ID NO:53,  NOV9
2.  SEQ ID NO:55,  Q9C0H6 KIAA1687 PROTEIN (FRAGMENT). homo sapiens. 6/2001
3.  SEQ ID NO:56,  Q9Y3J5 DA22D12.1. homo sapiens. 6/2001
4.  SEQ ID NO:57,  KHL1_HUMAN KELCH-LIKE PROTEIN 1. homo sapiens. 10/2000
5.  SEQ ID NO:58,  KHL1_MOUSE  KELCH-LIKE PROTEIN 1. mus musculus. 10/2000
6.  SEQ ID NO:59,  Q9H955 CDNA FLJ12995 FIS. homo sapiens. 6/2001


NOV9          1   EKAFVFPPATMSVSGKKEFDVKQILRLRWRWFSHP--FQGSTNTGSCLQQE----GYEHR 54
Q9C0H6        1   EKAFVFPPATMSVSGKKEFDVKQILRLRWRWFSHP--FQGSTNTGSCLQQE----GYEHR 54
Q9Y3J5        1   ------------------------------------------------------------ 1
KHL1_HUMAN    1   ----------MSGSGRKDFDVKHILRLRWKLFSHPSPSTGGPAGGGCLQQD-GSGSFEHW 49
KHL1_MOUSE    1   ----------MSGSGRKDFDVKHILRLRWKLFSHPSPASSSPAGGSCLQQDSGGGSFEHW 50
Q9H955        1   ------------------------------------------------------------ 1

NOV9         55   GTPVQGRLKSHSRD-------RNGLKKSNSPVHHNILAP----------VPGPAPAHQRA 97
Q9C0H6       55   GTPVQGRLKSHSRD-------RNGLKKSNSPVHHNILAP----------VPGPAPAHQRA 97
Q9Y3J5        1   ------------------------------------------------------------ 1
```





ProDom analysis indicates that the NOV9 polypeptide has 66 of 164 aa residues (40%) identical to, and 99 of 164 aa residues (60%) positive with, the 170 aa p36 (1) KELC_DROME – ring canal prptein (KELCH protein) repeat (prdm:36769, Expect = 2.0e-27); 64 of 191 aa residues (33%) identical to, and 98 of 191 aa residues (51%) positive with, the 265 aa p36 (36) SCRB(3) YC81(2) KELC(2) – protein repeat chromosome scruin EGF-like domain intergenic region cytoskeleton precursor (prdm:569, Expect = 2.9e-19); 50 of 201 aa residues (24%) identical to, and 99 of 201 aa residues (49%) positive with, the 263 aa p36 (3) VF03(2) VC13(1) -protein F3 C13, (prdm:9161, Expect = 8.5e-16); 41 of 116 aa residues (35%) identical to, and 65 of 116 aa residues (56%) positive with, the 220 aa p36 (30) BAC1(2) BCL6(2) Z151(2) – protein transcription nuclear DNA-binding regulation zinc-finger metal-binding zinc finger activator (prdm:716, Expect = 3.1e-12); and 29 of 115 aa residues (25%) identical to, and 57 of 115 aa residues (49%) positive with, the 148 aa p36 (4) VA55(2) VC02(2) – protein early A55 C2 (prdm:6493, Expect = 5.7e-07).

Pfam query for NOV9 indicates that NOV9 has high homology to two Interpro protein motifs, including the Kelch Kelch motif (Score=233.9, E-value=2.3e-66) and the BTB/POZ domain (Score=114.0, E-value=2.9e-30). PROSITE - software analysis indicates that NOV9 has one N-glycosylation site (Pattern-ID: ASN_glycosylation PS00001 (Interpro)); one cAMP- and cGMP-dependent protein kinase phosphorylation site (Pattern-ID: CAMP_PHOSPHO_SITE PS00004 (Interpro)); six Protein kinase C phosphorylation sites (Pattern-ID: PKC_PHOSPHO_SITE PS00005 (Interpro)); three Casein kinase II phosphorylation sites (Pattern-ID: CK2_PHOSPHO_SITE PS00006 (Interpro)); one Tyrosine kinase phosphorylation site (Pattern-ID: TYR_PHOSPHO_SITE PS00007 (Interpro)); eleven N-myristoylation sites (Pattern-ID: MYRISTYL PS00008 (Interpro)); and one Amidation site (Pattern-ID: AMIDATION PS00009 (Interpro)).

Table 9F lists the domain description from other domain analyses results against NOV9. This indicates that the NOV9 sequence has properties similar to those of other proteins known to contain this domain.

Table 9F. Domain Analysis of NOV9

```
Prodom                                                          Smallest Sum
                                                          High  Probability
Sequences producing High-scoring Segment Pairs:          Score  P(N)

prdm:36769 p36 (1) KELC_DROME - RING CANAL PROTEIN (KELC...   306  2.0e-27
prdm:569   p36 (36) SCRB(3) YC81(2) KELC(2)  - PROTEIN R...   231  5.8e-20
prdm:9161  p36 (3) VF03(2) VC13(1)  - PROTEIN F3 C13, 26...   199  8.5e-16
prdm:716   p36 (30) BAC1(2) BCL6(2) Z151(2)  - PROTEIN T...   166  3.1e-12
prdm:6493  p36 (4) VA55(2) VC02(2)  - PROTEIN EARLY A55 ...   117  5.7e-07

BLOCKS Protein Domain Analysis
AC#          Description                                    Strength  Score
BL00913B    0 Iron-containing alcohol dehydrogenases protei    1389   1043
BL00115S    0 Eukaryotic RNA polymerase II heptapeptide rep    1762   1040
BL00655C    0 Glycosyl hydrolases family 6 proteins.           1384   1037
BL01092Q    0 Adenylate cyclases class-I proteins.             1997   1035
BL01066D    0 Uncharacterized protein family UPF0015 protei    1584   1029

BLOCKS Protein Domain Analysis                          NOV9 aa position
Pattern-ID: ASN_GLYCOSYLATION PS00001 (Interpro)                   2
Pattern-DE: N-glycosylation site, Pattern: N[^P][ST][^P]

Pattern-ID: CAMP_PHOSPHO_SITE PS00004 (Interpro)                 275
Pattern-DE: cAMP- and cGMP-dependent protein kinase phosphorylation site
Pattern:    [RK]{2}.[ST]

Pattern-ID: PKC_PHOSPHO_SITE PS00005 (Interpro)       19, 269, 362, 409, 445, 455
Pattern-DE: Protein kinase C phosphorylation site
Pattern:    [ST].[RK]

Pattern-ID: CK2_PHOSPHO_SITE PS00006 (Interpro)          4, 140, 295
Pattern-DE: Casein kinase II phosphorylation site
Pattern:    [ST].{2}[DE]

Pattern-ID: TYR_PHOSPHO_SITE PS00007 (Interpro)                  249
Pattern-DE: Tyrosine kinase phosphorylation site
Pattern:    [RK].{2,3}[DE].{2,3}Y

Pattern-ID: MYRISTYL PS00008 (Interpro)                78,  216, 280, 286, 311, 333,
Pattern-DE: N-myristoylation site                        366, 380, 427, 460, 527
Pattern:    G[^EDRKHPFYW].{2}[STAGCN][^P]

Pattern-ID: AMIDATION PS00009 (Interpro)                         314
Pattern-DE: Amidation site, Pattern: .G[RK][RK]
```

Other BLAST results include sequences from the Patp database, which is a proprietary database that contains sequences published in patents and patent publications. BLASTP analysis of the patp database shows that NOV9 has 569 of 569 aa residues (100%) identical to, and 569 of 569 aa residues (100%) positive with, the 569 aa Human protein sequence SEQ ID NO:14569 (patp:AAB94214, Expect = 2.8e-314); 411 of 411 aa residues (100%) identical to, and 411 of 411 aa residues (100%) positive with, the 411 aa Human protein sequence SEQ ID NO:14985 (patp:AAB94406, Expect = 7.3e-229); 381 of 508 aa residues (75%) identical to, and 439 of 508 aa residues (86%) positive with, the 508 aa Human protein sequence SEQ ID NO:13220 (patp:AAB93678, Expect = 9.8e-218); 380 of 508 aa residues (74%) identical to, and 438 of 508 aa residues (86%) positive with, the 508 aa Human protein sequence SEQ ID NO:12231 (patp:AAB93233, Expect = 8.8e-217); and 242 of 554 aa residues (43%) identical to, and 349 of

554 aa residues (62%) positive with. the 609 aa Human protein sequence SEQ ID NO:11635 (patp:AAB92953, Expect = 2.9e-122). Patp results include those listed in Table 9GF.

Table 9G. Patp alignments of NOV9

Sequences producing High-scoring Segment Pairs:	High Score	Smallest Sum Prob. P(N)
patp:AAB94214 Human protein sequence SEQ ID NO:14569 - Ho...	3015	2.8e-314
patp:AAB94406 Human protein sequence SEQ ID NO:14985 - Ho...	2209	7.3e-229
patp:AAB93678 Human protein sequence SEQ ID NO:13220 - Ho...	2104	9.8e-218
patp:AAB93233 Human protein sequence SEQ ID NO:12231 - Ho...	2095	8.8e-217
patp:AAB92953 Human protein sequence SEQ ID NO:11635 - Ho...	1203	2.9e-122

The kelch motif was discovered as a sixfold tandem element in the sequence of the Drosophila kelch ORF1 protein. The repeated kelch motifs predict a conserved tertiary structure, a beta-propeller. This module appears in many different polypeptide contexts and contains multiple potential protein-protein contact sites. Members of this growing superfamily are present throughout the cell and extracellularly and have diverse activities.

The Drosophila kelch protein is a structural component of ring canals and is required for oocyte maturation. Recently, a new human homologue of kelch, KLHL3, was cloned. At the amino acid level, KLHL3 shares 77% similarity with Drosophila kelch and 89% similarity with Mayven (KLHL2), another human kelch homolog. Like kelch and KLHL2, the KLHL3 protein contains a poxvirus and zinc finger domain at the N-terminus and six tandem repeats (kelch repeats) at the C-terminus. Various KLHL3 isoforms result from alternative promoter usage, alternative polyadenylation sites and alternative splicing. The KLHL3 gene is mapped to human chromosome 5, band q31, contains 17 exons, and spans approximately 120 kb of genomic DNA. KLHL3 maps within the smallest commonly deleted segment in myeloid leukemias characterized by a deletion of 5q; however, no inactivating mutations of KLHL3 were detected in malignant myeloid disorders with loss of 5q.

The disclosed NOV9 nucleic acid encoding a Kelch -like protein includes the nucleic acid whose sequence is provided in Table 9A, or a fragment thereof. The invention also includes a mutant or variant nucleic acid any of whose bases may be changed from the corresponding base shown in Table 9A while still encoding a protein that maintains its Kelch -like activities and physiological functions, or a fragment of such a nucleic acid. The invention further includes nucleic acids whose sequences are complementary to those just described, including nucleic acid fragments that are complementary to any of the nucleic acids just described. The invention additionally includes nucleic acids or nucleic acid fragments, or complements thereto, whose structures include chemical modifications. Such modifications include, by way of nonlimiting

example, modified bases, and nucleic acids whose sugar phosphate backbones are modified or derivatized. These modifications are carried out at least in part to enhance the chemical stability of the modified nucleic acid, such that they may be used. for example, as antisense binding nucleic acids in therapeutic applications in a subject.

The disclosed NOV9 nucleic acid is useful as a marker for Menkes disease, myoglobinuria/hemolysis due to PGK deficiency. Wieacker-Wolff syndrome and/or other diseases/disorders.

Based on the tissues in which NOV9 is most highly expressed; including uterus, brain breast, and stomach; specific uses include developing products for the diagnosis or treatment of a variety of diseases and disorders. Additional disease indications and tissue expression for NOV9 is presented in Example 2.

The disclosed NOV9 protein of the invention includes the Kelch -like protein whose sequence is provided in Table 9B. The invention also includes a mutant or variant protein any of whose residues may be changed from the corresponding residue shown in Table 9B while still encoding a protein that maintains its Kelch -like activities and physiological functions, or a functional fragment thereof.

The invention further encompasses antibodies and antibody fragments, such as F_{ab} or $(F_{ab})_2$ that bind immunospecifically to any of the proteins of the invention.

The above defined information for this invention suggests that this Kelch -like protein (NOV9) may function as a member of a "Kelch family". Therefore, the NOV9 nucleic acids and proteins identified here may be useful in potential therapeutic applications implicated in (but not limited to) various pathologies and disorders as indicated below. The potential therapeutic applications for this invention include, but are not limited to: leukemia research tools, for all tissues and cell types composing (but not limited to) those defined here.

The NOV9 nucleic acids and proteins of the invention are useful in potential therapeutic applications implicated in cancer including but not limited to leukemias and/or other pathologies and disorders. For example, a cDNA encoding the Kelch -like protein (NOV9) may be useful in disease therapy for Menkes disease, myoglobinuria/hemolysis due to PGK deficiency, and Wieacker-Wolff syndrome, and the Kelch -like protein (NOV9) may be useful when administered to a subject in need thereof. By way of nonlimiting example, the compositions of the present invention will have efficacy for treatment of patients suffering from neurological disorders including but not limited to Menkes disease. The NOV9 nucleic acid encoding Kelch -like protein, and the Kelch -like protein of the invention, or fragments thereof, may further be useful in diagnostic applications, wherein the presence or amount of the nucleic acid or the protein are to be assessed.

NOV9 nucleic acids and polypeptides are further useful in the generation of antibodies that bind immuno-specifically to the novel NOV9 substances for use in therapeutic or diagnostic methods. These antibodies may be generated according to methods known in the art, using prediction from hydrophobicity charts, as described in the "Anti-NOVX Antibodies" section below. The disclosed NOV9 protein has multiple hydrophilic regions, each of which can be used as an immunogen. In one embodiment, a contemplated NOV9 epitope is from about amino acids 1 to 40. In another embodiment, a NOV9 epitope is from about amino acids 60-95. In additional embodiments, NOV9 epitopes are from about amino acids 130 to 220, from about amino acids 240-320, from about amino acids 330 to 370, from about amino acids 380 to 415, from about amino acids 425 to 460, from about amino acids 470 to 510 and from about amino acids 520 to 569. These novel proteins can be used in assay systems for functional analysis of various human disorders, which will help in understanding of pathology of the disease and development of new drug targets for various disorders.

NOV10

NOV10 includes three novel Type IIIb plasma membrane-like proteins disclosed below. The disclosed NOV10 proteins have been named NOV10a, NOV10b and NOV10c.

NOV10a

A disclosed NOV10a nucleic acid of 1339 nucleotides (also referred to as 100340173; 1373975; 1373976; 1373977 and1373978) encoding a novel hypothetical Y305_SYNY3 22.2 kDa prrotein SLR0305-like protein/ Type IIIb plasma membrane-like proteins is shown in Table 10A. An open reading frame was identified beginning with an ATG initiation codon at nucleotides 367-369 and ending with a TGA codon at nucleotides 925-927. A putative untranslated region upstream from the initiation codon and downstream from the termination codon is underlined in Table 10A, and the start and stop codons are in bold letters.

Table 10A. NOV10a nucleotide sequence (SEQ ID NO:60).

CACGGTCCGCCCAGAGGCTTCGGAGCTGCCGGAGCCGGGCGGGGCCTTGGCGGGCGGCCCCGGGAGTGGCGGCGGCGGCGTG
GTGGTCGGCGTGGCTGAGGTGAGAAACTGGCGCTGCGGCTGCCTCGGAGCACCTGTTGGTGCCGGAGCCTCGTGCTGGTCTG
CGTGTTGGCCGCCCTGTGCTTCGCTTCCCTGGCCCTGGTCCCGCCGCTACCTTCACCACCTCCTGCTGTGGTGGAGAGCCTT
GACTCGCTGCTGGGGGTCCTGCTCTTCGTCGTGGCTCATCGTGTTCTTTCCCCTGCGGCCGGGGCTACATCGTGCTCA
ACGTGGCCGCTGGCTACCTGTACGGCTTCGTGCTGGGCATGGGTCTGATGATGGTGGGCGTCCTCATCGGCACCTTCATCGC
CCATGTGGTCTGCAAGCGGCTCCTCACCGCCTGGGTGGCCGCCAGGATCCAGAGCAGCGAGAAGCTGAGCGCCGTTATTCGC
GTAGTGGAGGGAGGAAGCGGCCTGAAAGTGGTGGCGCTGGCCAGACTGACACCCATACCTTTTGGGCTTCAGAATGCAGTGT
TTTCGATTACTGATCTCTCATTACCCAACTATCTGATGGCATCTTCGGTTGGACTGCTTCCTACCCAGCTTCTGAATTCTTA
CTTGGGTACCACCCTGCGGACAATGGAAGATGTCATTGCAGAACAGAGTGTTAGTGGATATTTTGTTTTTTGTTTACAGATT
ATTATAAGTATAGGCCTCATGTTTTATGTAGTTCATCGAGCTCAAGTGGAATTGAATGCAGCTATTGTAGCTTGTGAAATGG
AACTGAAATCTTCTCTGGTTAAAGGCAATCAACCAAATACCAGTGGCTCTTCATTCTACAACAAGAGGACCCTAACATTTTC
TGGAGGTGGAATCAATGTTGTATGATTCTAATGAGATACGTGATTGTCAAGAGCCTAGTGTGCTATCTAAGGTCTAGCAGTC
ACTTCACTAGTGGGCAGAGACAAGTTCTAATTGTATTACAGCACAAACAAAACTGACTAGTTTTTAAATTGCACAATTTTTT
TTTTTTTAAGCAAGAATCATTTTCTGGGTATGTAAGTGTAAATGTAGATGCAAATTTGGCTGCACCTCTTTATCATGCCTGT
ATTGGCCTATAGGTCTGCACTTTAGTGTTTTTTAATTGTTTTATTTCTGTGTATTTACGAACAGAGAAATAACTCAAATATT

ATTTCTGCTTAGTGTCTTTATTTATAAAGCCATGAGTAGTTTGTATGCATCTTTCCTACTTGTAAAGATGAGTAAAAGTAT
GCAGTTTTAAATTTAAAAAAAAAAAAA

A disclosed NOV10a polypeptide (SEQ ID NO:61) encoded by SEQ ID NO:60 has 186 amino acid residues and is presented in Table 10B using the one-letter amino acid code. SignalP, Psort and/or Hydropathy results predict that NOV10 has a signal peptide and is likely to be localized endoplasmic reticulum (membrane) with a certainty of 0.6850. In alternative embodiments, the NOV10a protein localizes to the plasma membrane with a certainty of 0.6400; a Golgi body with a certainty of 0.4600; or the endoplasmic reticulum (lumen) with a certainty of 0.1000. The most likely cleavage site for a NOV10a peptide is between amino acids 19 and 20, at: VVC-KR. NOV10a has a molecular weight of 19946.3 Daltons.

Table 10B. Encoded NOV10a protein sequence (SEQ ID NO:61).

MGLMMVGVLIGTFIAHVVCKRLLTAWVAARIQSSEKLSAVIRVVEGGSGLKVVALARLTPIPFGLQNAVFSITDLSLPNVLM
ASSVGLLPTQLLNSYLGTTLRTMEDVIAEQSVSGYFVFCLQIIISIGLMFYVVHRAQVELNAAIVACEMELKSSLVKGNQPN
TSGSSFYNKRTLTFSGGGINVV

NOV10b

A disclosed NOV10 nucleic acid of 512 nucleotides (also referred to as CG56409-02) encoding a novel hypothetical 22.2 kDa prtotein SLR0305-like, Type IIIb Plasma Membrane-like, protein is shown in Table 10C. The sequence was derived by laboratory cloning of cDNA fragments and by *in silico* prediction of the sequence. An open reading frame was identified beginning with an ATG initiation codon at nucleotides 108-110 and ending with a TGA codon at nucleotides 510-512. A putative untranslated region upstream from the initiation codon is underlined in Table 10C, and the start and stop codons are in bold letters.

Table 10C. NOV10b nucleotide sequence (SEQ ID NO:62).

GGGTCCTGCTCTTCGTCGTGGGCTTCATCGTGGTCTCTTTCCCCTGCGGCTGGGGCTACATCGTGCTCAACGTGGCCG
CTGGCTACCTGTACGGCTTCGTGCTGGGCATGGGTCTGATGATGGTGGGCGTCCTCATCGGCACCTTCATCGCCCATG
TGGTCTGCAAGCGGCTCCTCACCGCCTGGGTGGCCGCCAGGATCCAGAGCAGCGAGAAGCTGAGCGCGGTTATTCGCG
TAGTGGAGGGAGGAAGCGGCCTGAAAGTGGTGGCGCTGGCCAGACTGACACCCATACCTTTTGGGCTTCAGAATGCAG
TGTTTTCGATTATTATAAGTATAGGCCTCATGTTTTATGTAGTTCATCGAGCTCAAGTGGAATTGAATGCAGCTATTG
TAGCTTGTGAAATGGAACTGAAATCTTCTCTGGTTAAAGGCAATCAACCAAATACCAGTGGCTCTTCATTCTACAACA
AGAGGACCCTAACATTTTCTGGAGGTGGAATCAATGTTGTATGA

A disclosed NOV10b polypeptide (SEQ ID NO:63) encoded by SEQ ID NO:62 has 134 amino acid residues and is presented in Table 10D using the one-letter amino acid code. SignalP, Psort and/or Hydropathy results predict that NOV10b has a signal peptide, cleavage site

and localization results analogous to those listed for NOV10a and NOV10c. Additional software analysis suggests that NOV10b has an INTEGRAL likelihood of -6.74 for a predicted transmembrane region at aa3 - aa19 (1 - 20) and an INTEGRAL likelihood of -5.47 for a predicted transmembrane region at aa68 - aa84 (63 - 86), and that it is likely a Type IIIb membrane protein (Nexo Ccyt). NOV10b has a molecular weight of 14249.2 Daltons.

Table 10D. Encoded NOV10b protein sequence (SEQ ID NO:63).

MGLMMVGVLIGTFIAHVVCXRLLTAWVAARIQSSEKLSAVIRVVEGGSGLKVVALARLTPIPPGLQNAVFSIIISIGLMFYV
VHRAQVELNAAIVACEMELKSSLVKGNQPNTSGSSFYNKRTLTFSGGGINVV

NOV10c

A disclosed NOV10c nucleic acid of 1339 nucleotides (also referred to as CG56409-03) encoding a novel hypothetical 22.2 kDa prtotein SLR0305-like protein is shown in Table 10E. An open reading frame was identified beginning with an ATG initiation codon at nucleotides 1-3 and ending with a TGA codon at nucleotides 649-651. A putative untranslated region downstream from the termination codon is underlined in Table 10E, and the start and stop codons are in bold letters.

Table 10E. NOV10c nucleotide sequence (SEQ ID NO:64).

ATGGGCTTCATCGTGGTCTCTTTCCCCTGCGGCTGGGGCTACATCGTGCTCAACGTGGCCGCTGGCTACCTGTACGGC
TTCGTGCTGGGCATGGGTCTGATGATGGTGGGCGTCCTCATCGGCACCTTCATCGCCCATGTGGTCTGCAAGCGGCTC
CTCACCGCCTGGGTGGCCGCCAGGATCCAGAGCAGCGAGAAGCTGAGCGCGGTTATTCGCGTAGTGGAGGGAGGAAGC
GGCCTGAAAGTGGTGGCGCTGGCCAGACTGACACCCATACCTTTTGGGCTTCAGAATGCGGTGTTTTCGATTACTGAT
CTCTCATTACCCAACTATCTGATGGCATCTTCGGTTGGACTGCTTCCTACCCAGCTTCTGAATTCTTACTTGGGTACC
ACCCTGCGGACAATGGAAGATGTCATTGCAGAACAGAGTGTTAGTGGATATTTTGTTTTTTGTTTACAGATTATTATA
AGTATAGGCCTCATGTTTTATGTAGTTCATCGAGCTCAAGTGGAATTGAATGCAGCTATTGTAGCTTGTGAAATGGAA
CTGAAATCTTCTCTGGTTAAAGGCAATCAACCAAATACCAGTGGCTCTTCATTCTACAACAAGAGGACCCTAACATTT
TCTGGAGGTGGAATCAATGTTGTA**TGA**TTCTAATGAGATACGTGATTGTTAAGAGCCTAGTGTGTA

A disclosed NOV10c polypeptide (SEQ ID NO:65) encoded by SEQ ID NO:64 has 216 amino acid residues and is presented in Table 10F using the one-letter amino acid code. SignalP, Psort and/or Hydropathy results predict that NOV10c has a signal peptide, cleavage site and localization results analogous to those listed for NOV10a and NOV10b. Additional software analysis suggests that NOV10c has an INTEGRAL likelihood of -8.12 for a predicted transmembrane region at aa149 - aa165 (142 - 167) and an INTEGRAL likelihood of -6.74 for a predicted transmembrane region at aa33 - aa49 (22 - 50), and that it is likely a Type IIIb membrane protein (Nexo Ccyt). The most likely cleavage site for a NOV10c peptide is between amino acids 49 and 50, at: VVC-KR. NOV10c has a molecular weight of 23141 Daltons.

Table 10F. Encoded NOV10c protein sequence (SEQ ID NO:65).

```
MGFIVVSFPCGWGYIVLNVAAGYLYGFVLGMGLMMVGVLIGTFIAHVVCKRLLTAWVAARIQSSEKLSAVIRVVEGGSGLKV
VALARLTPIPPGLQNAVFSITDLSLPNYLMASSVGLLPTQLLNSYLGTTLRTMEDVIAEQSVSGYFVFCLQIIISIGLMFYV
VHRAQVELNAAIVACEMELKSSLVKGNQPNTSGSSFYNKRTLTFSGGGINVV
```

NOV10a, NOV10b and NOV10c polypeptides are related to each other as shown in the ClustalW alignment in Table 10G.

Table 10G: ClustalW of NOV10 Variants

```
NOV10a  ------------------------------MGLMMVGVLIGTFIAHVVCK 20
NOV10b  ------------------------------MGLMMVGVLIGTFIAHVVCK 20
NOV10c  MGFIVVSFPCGWGYIVLNVAAGYLYGFVLGMGLMMVGVLIGTFIAHVVCK 50

NOV10a  RLLTAWVAARIQSSEKLSAVIRVVEGGSGLKVVALARLTPIPPGLQNAVF 70
NOV10b  RLLTAWVAARIQSSEKLSAVIRVVEGGSGLKVVALARLTPIPPGLQNAVF 70
NOV10c  RLLTAWVAARIQSSEKLSAVIRVVEGGSGLKVVALARLTPIPPGLQNAVF 100

NOV10a  SITDLSLPNYLMASSVGLLPTQLLNSYLGTTLRTMEDVIAEQSVSGYFVF 120
NOV10b  S------------------------------------------------- 71
NOV10c  SITDLSLPNYLMASSVGLLPTQLLNSYLGTTLRTMEDVIAEQSVSGYFVF 150

NOV10a  CLQIIISIGLMFYVVHRAQVELNAAIVACEMELKSSLVKGNQPNTSGSSF 170
NOV10b  ---IIISIGLMFYVVHRAQVELNAAIVACEMELKSSLVKGNQPNTSGSSF 118
NOV10c  CLQIIISIGLMFYVVHRAQVELNAAIVACEMELKSSLVKGNQPNTSGSSF 200

NOV10a  YNKRTLTFSGGGINVV 186
NOV10b  YNKRTLTFSGGGINVV 134
NOV10c  YNKRTLTFSGGGINVV 216
```

Additional NOV10 SNP and coding variant sequences are described in Example 3.

In a search of sequence databases, it was found, for example, that the NOV10b nucleic acid sequence has 156 of 245 bases (63%) identical to a gb:GenBank-ID:MFU72744|acc:U72744.1 mRNA from Mycobacterium fortuitum (Mycobacterium fortuitum nitrite extrusion protein gene, complete cds). The full NOV10b amino acid sequence was found to have 29 of 80 amino acid residues (36%) identical to, and 45 of 80 amino acid residues (56%) similar to, the 209 amino acid residue ptnr:SwissProt-ACC:Q55909 protein from Synechocystis sp. (strain PCC 6803) (hypothetical 22.2 kDa protein SLR0305). In a search of sequence databases, it was found, for example, that the NOV10c nucleic acid sequence has 156 of 245 bases (63%) identical to a gb:GenBank-ID:MFU72744|acc:U72744.1 mRNA from Mycobacterium fortuitum (Mycobacterium fortuitum nitrite extrusion protein gene, complete cds). The full NOV10c amino acid sequence of the protein of the invention was found to have 52 of 170 amino acid residues (30%) identical to, and 96 of 170 amino acid residues (56%) similar to, the 209 amino acid residue ptnr: SwissProt -ACC:Q55909 protein from Synechocystis sp. (strain PCC 6803) (hypothetical 22.2 kDa protein SLR0305).

In an additional search of public protein databases, the NOV10a amino acid sequences have homology to the amino acid sequences shown in the BLASTP data listed in Table 10H. Public amino acid databases include the GenBank databases, SwissProt, PDB and PIR.

Table 10H. BLAST results for NOV10a

Gene Index/ Identifier	Protein/ Organism	Length (aa)	Identity (%)	Positives (%)	Expect
Y305_SYNY3; D64005; BAA10672.1; Q55909	HYPOTHETICAL 22.2 KDA PROTEIN SLR0305. synechocystis sp. (strain pcc 6803). 11/1997	209	46/154 (30%)	86/154, (56%)	3e-12
Q9VNR8; AE003598; AAF51854.2	CG11367 PROTEIN. drosophila melanogaster. 3/2001	834	28/81 (35%)	56/81, (69%)	6e-10
Q9ZVS7; AC005278; AAC72122.1	F15K9.14. arabidopsis thaliana. 5/1999	269	41/153 (27%)	82/153, (54%)	7e-09
Q9RPT3; AF148265; AAD55929.1	HYPOTHETICAL TRANSMEMBRANE PROTEIN. uncultured bacterium ah1. 5/2000	225	40/144 (28%)	73/144, (51%)	2e-05

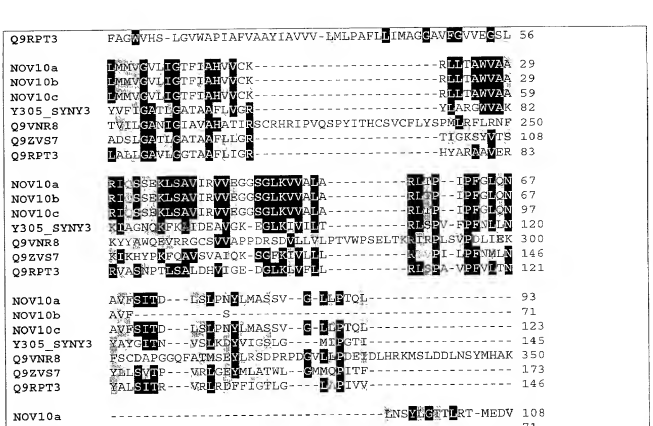
The homology of these and other sequences is shown graphically in the ClustalW analysis shown in Table 10I. In the ClustalW alignment of the NOV10 proteins, as well as all other ClustalW analyses herein, the black outlined amino acid residues indicate regions of conserved sequence (*i.e.*, regions that may be required to preserve structural or functional properties), whereas non-highlighted amino acid residues are less conserved and can potentially be mutated to a much broader extent without altering protein structure or function.

Table 10I. ClustalW Analysis of NOV10

```
1)  NOV10a   (SEQ ID NO:61)
2)  NOV10b   (SEQ ID NO:63)
3)  NOV10c   (SEQ ID NO:65)
4)  Y305_SYNY3  (SEQ ID NO:66)
5)  Q9VNR8 (partial sequence) (SEQ ID NO:67)
6)  Q9ZVS7  (SEQ ID NO:68)
7)  Q9RPT3  (SEQ ID NO:69)

NOV10a      ------------------------------------------------- 1
NOV10b      ------------------------------------------------- 1
NOV10c      ------------------------------------------------- 1
Y305_SYNY3  ----------------------------------MA-----DYLLN 7
Q9VNR8      ...HNRKRNSCWGRAHSFLTRNWYLGCLVPATILGALVFIGWATRDYARQ 150
Q9ZVS7      ------------MSFTPSTFRIAISLLLLVAIVSAVIFL-----PKLKD 32
Q9RPT3      ----------------------------------MVS-----PWLPS 8

NOV10a      ----------------------------------------------MG 2
NOV10b      ----------------------------------------------MG 2
NOV10c      ----------------MGFIVVSFP-CGWGYIVLNVAASYLYCFVLSMG 32
Y305_SYNY3  ALQWTDG-LGTWAAIAFMLLYTVATV-VFLPGSTLTGAGVVEGVILCSI 55
Q9VNR8      LLFWTEMQNAWITFAVYMGLFALVSFPVVVGYFWLLITAGYLDGCLRGWM 200
Q9ZVS7      FLLWTKEDLGPFGPLALALAMIPLTI-WAVPASVWLTLGGGYLDGLPVGFW 81
```



The presence of identifiable domains in NOV10a, and to NOV10b and NOV10c in analogous regions, was determined. DOMAIN results for NOV10 as disclosed in Tables 10J, were collected from the Conserved Domain Database (CDD) with Reverse Position Specific

BLAST analyses. This BLAST analysis software samples domains found in the Smart and Pfam collections.

ProDom analysis of NOV10a shows homology to various domains. Specifically, NOV10a has 32 of 124 aa residues (25%) identical to, and 67 of 124 aa residues (54%) positive with, the 208 aa p36 (7) protein transmembrane intergenic region CY20H10.06C SLR0305 CY277.13C XTHA-GDHA NUCB-AROD DNAI-THRS (prdm:3727, Expect = 2.7e-08); 14 of 36 aa residues (38%) identical to, and 21 of 36 aa residues (58%) positive with, the 68 aa p36 (1) NU2M_HANWI - NADH-ubiquinone oxidoreductase chain 2 (EC 1.6.5.3)(prdm:21748, Expect = 0.27); 13 of 30 aa residues (43%) identical to, and 18 of 30 aa residues (60%) positive with, the 41 aa p36 (1) SODE_DIRIM – extracellular superoxide dismutase precursor (CU-ZN) (EC 1.15.1.1) (EC-SOD)(prdm:27499, Expect = 0.27); 15 of 54 (27%) identical to, and 23 of 54 (42%) positive with, the 69 aa p36 (1) RL37_TETTH – ribosomal protein L37 (P1 TYPE) (prdm:21871, Expect = 0.74); and 14 of 31 aa residues (45%) identical to, and 20 of 31 aa residues (64%) positive with, the 158 aa p36 (1) YIK5_YEAST - hypothetical 78.0 KD protein in MOB1-SGA1 intergenic region (prdm:55957, Expect = 1.3). Table 10J lists various domain description from domain software analysis results against NOV10. This indicates that the NOV10 sequence has properties similar to those of other proteins known to contain this domain.

Table 10J. Domain Analysis of NOV10

PFAM HMM Domain Analysis of NOV10

Model	Domain	seq-f	seq-t	hmm-f	hmm-t	score	E-value
[no hits above thresholds]							

ProDom analysis

	High Score	Smallest Sum Probability P(N)
Sequences producing High-scoring Segment Pairs:		
prdm:3727 p36 (7) - PROTEIN TRANSMEMBRANE INTERGENIC R...	129	2.7e-08
prdm:21748 p36 (1) NU2M_HANWI - NADH-UBIQUINONE OXIDORED...	58	0.23
prdm:27499 p36 (1) SODE_DIRIM - EXTRACELLULAR SUPEROXIDE...	58	0.23
prdm:21871 p36 (1) RL37_TETTH - RIBOSOMAL PROTEIN L37 (P...	54	0.52
prdm:55957 p36 (1) YIK5_YEAST - HYPOTHETICAL 78.0 KD PRO...	68	0.73

BLOCKS Protein Domain Analysis

AC#	Description	Strength	Score
BL00495E	0 Apple domain proteins.	1844	1049
BL00505C	0 Phosphoenolpyruvate carboxykinase (GTP) prote	1787	1019
BL00853C	0 Beta-eliminating lyases pyridoxal-phosphate a	1544	1017
BL01235B	0 Uncharacterized protein family UPF0019 protei	2114	1016

PROSITE Analysis

Pattern-ID: ASN_GLYCOSYLATION PS00001 (Interpro)
one N-glycosylation site

Pattern-ID: GLYCOSAMINOGLYCAN PS00002 (Interpro)
one Glycosaminoglycan attachment site

Pattern-ID: PKC_PHOSPHO_SITE PS00005 (Interpro)
two Protein kinase C phosphorylation sites

```
Pattern-ID: CK2_PHOSPHO_SITE PS00006 (Interpro)
two Casein kinase II phosphorylation sites

Pattern-ID: MYRISTYL PS00008 (Interpro)
five N-myristoylation sites
```

Other BLAST results include sequences from the Patp database, which is a proprietary database that contains sequences published in patents and patent publications. In a BLASTP analysis of the patp database, NOV10 was found to have 93 of 102 aa residues (91%) identical to, and 95 of 102 aa residues (93%) positive with, the 111 aa Human prostate cancer antigen protein sequence SEQ ID NO:1245 (patp:AAB56667, Expect = 3.0e-42); 45 of 144 aa residues (31%) identical to, and 80 of 144 aa residues (55%) positive with, the 280 aa Arabidopsis thaliana protein fragment SEQ ID NO: 12140 (patp:AAG12863, Expect = 1.6e-12); 39 of 130 aa residues (30%) identical to, and 66 of 130 aa residues (50%) positive with, the 174 aa Arabidopsis thaliana protein fragment SEQ ID NO: 64446 (patp:AAG50824, Expect = 3.0e-06); 39 of 130 aa residues (30%) identical to, and 66 of 130 aa residues (50%) positive with, the 204 aa Arabidopsis thaliana protein fragment SEQ ID NO: 37254 (patp:AAG31071, Expect = 9.5e-06); and 39 of 130 aa residues (30%) identical to, and 66 of 130 aa residues (50%) positive with, the 204 aa Arabidopsis thaliana protein fragment SEQ ID NO: 64445 (patp:AAG50823, Expect = 9.5e-06). Patp results include those listed in Table 10K.

Table 10K. Patp alignments of NOV10

Sequences producing High-scoring Segment Pairs:	High Score	Smallest Sum Prob. P(N)
patp:AAB56667 Human prostate cancer antigen protein seque...	448	3.0e-42
patp:AAG12863 Arabidopsis thaliana protein fragment SEQ I...	169	1.6e-12
patp:AAG50824 Arabidopsis thaliana protein fragment SEQ I...	118	3.0e-06
patp:AAG31071 Arabidopsis thaliana protein fragment SEQ I...	118	9.5e-06
patp:AAG50823 Arabidopsis thaliana protein fragment SEQ I...	118	9.5e-06

The Type IIIb Plasma Membrane-like NOV10 disclosed in this invention maps to chromosome 8q13 and 8q21. This assignment was made using mapping information associated with genomic clones, public genes and ESTs sharing sequence identity with the disclosed sequence and CuraGen Corporation's Electronic Northern bioinformatic tool.

The disclosed NOV10 nucleic acid encoding a novel hypothetical 22.2 kDa prtotein SLR0305-like protein includes the nucleic acid whose sequence is provided in Table 10A, or a fragment thereof. The invention also includes a mutant or variant nucleic acid any of whose bases may be changed from the corresponding base shown in Table 10A while still encoding a protein that maintains its novel hypothetical 22.2 kDa prtotein SLR0305-like protein activities

and physiological functions, or a fragment of such a nucleic acid. The invention further includes nucleic acids whose sequences are complementary to those just described, including nucleic acid fragments that are complementary to any of the nucleic acids just described. The invention additionally includes nucleic acids or nucleic acid fragments, or complements thereto. whose structures include chemical modifications. Such modifications include, by way of nonlimiting example, modified bases, and nucleic acids whose sugar phosphate backbones are modified or derivatized. These modifications are carried out at least in part to enhance the chemical stability of the modified nucleic acid, such that they may be used, for example, as antisense binding nucleic acids in therapeutic applications in a subject. In the mutant or variant nucleic acids, and their complements, up to about 37 % percent of the bases may be so changed.

The disclosed NOV10 protein of the invention includes the novel hypothetical 22.2 kDa prtotein SLR0305-like protein whose sequence is provided in Table 10B. The invention also includes a mutant or variant protein any of whose residues may be changed from the corresponding residue shown in Table 10B while still encoding a protein that maintains its novel hypothetical 22.2 kDa prtotein SLR0305-like activities and physiological functions, or a functional fragment thereof. In the mutant or variant protein, up to about 64 % percent of the residues may be so changed.

The Type IIIb Plasma Membrane-like NOV10 gene disclosed in this invention is expressed in at least in peripheral blood tissues. Expression information was derived from the tissue sources of the sequences that were included in the derivation of the sequence, as provided in Example 1.

The invention further encompasses antibodies and antibody fragments, such as F_{ab} or $(F_{ab})_2$, that bind immunospecifically to any of the proteins of the invention.

The above defined information for this invention suggests that this novel hypothetical 22.2 kDa prtotein SLR0305-like protein (NOV10) may function as a member of a "Type IIIb plasma membrane-like protein family". Therefore, the NOV10 nucleic acids and proteins identified here may be useful in potential therapeutic applications implicated in (but not limited to) various pathologies and disorders as indicated below. The potential therapeutic applications for this invention include, but are not limited to: Type IIIb plasma membrane-related research tools, for all tissues and cell types composing (but not limited to) those defined herein.

The NOV10 nucleic acids and proteins of the invention are useful in potential therapeutic applications implicated in cancer including but not limited to disorders such as neural, immune, muscular, reproductive, gastrointestinal, pulmonary, cardiovascular, renal, and proliferative disorders, wounds, and infectious diseases. and/or other pathologies and disorders. For example, a cDNA encoding the SLR0305-like NOV10 protein may be useful in gene and protein therapy,

and the SLR0305-like protein (NOV10) may be useful when administered to a subject in need thereof. By way of nonlimiting example, the compositions of the present invention will have efficacy for treatment of patients suffering from Type IIIb plasma membrane-related disorders including but not limited to those described in the Examples. The NOV10 nucleic acid encoding the SLR0305-like protein, and the SLR0305-like protein of the invention, or fragments thereof, may further be useful in diagnostic applications, wherein the presence or amount of the nucleic acid or the protein are to be assessed.

The protein similarity information, expression pattern, cellular localization, and map location for the protein and nucleic acid disclosed herein suggest that this Type IIIb Plasma Membrane-like NOV10 protein may have important structural and/or physiological functions characteristic of the Type IIIb Plasma Membrane family.

The NOV10 nucleic acids and proteins of the invention have applications in the diagnosis and/or treatment of various diseases and disorders. For example, the NOV10 compositions of the present invention will have efficacy for the treatment of patients suffering from: ACTH deficiency; familial febrile convulsions 1; Duane syndrome; congenital Adrenal hyperplasia due to 11-beta-hydroxylase deficiency; glucocorticoid-remediable Aldosteronism; congenital Hypoaldosteronism due to CMO I deficiency; congenital Hypoaldosteronism due to CMO II deficiency; Nijmegen breakage syndrome; susceptibility to Low renin hypertension; Anemia, Ataxia-telangiectasia, Autoimmume disease, Immunodeficiencies as well as other diseases, disorders and conditions.

These materials are further useful in the generation of antibodies that bind immunospecifically to the novel substances of the invention for use in diagnostic and/or therapeutic methods.

NOV10 nucleic acids and polypeptides are further useful in the generation of antibodies that bind immuno-specifically to the novel NOV10 substances for use in therapeutic or diagnostic methods. These antibodies may be generated according to methods known in the art, using prediction from hydrophobicity charts, as described in the "Anti-NOVX Antibodies" section below. The disclosed NOV10a protein has multiple hydrophilic regions, each of which can be used as an immunogen. In one embodiment, a contemplated NOV10a epitope is from about amino acids 18 to 25. In another embodiment, a NOV10 epitope is from about amino acids 30 to 50. In additional embodiments, NOV10a epitopes are from about amino acids 100 to 120 and from about amino acids 135 to 186. In another embodiment, a contemplated NOV10b epitope is from about amino acids 25 to 45 and from about amino acids 100 to 134. In a further embodiment, a contemplated NOV10c epitope is from about amino acids 50 to 75, from about amino acids 120 to 145 and from about amino acids 180 to 216. These novel NOV10 proteins

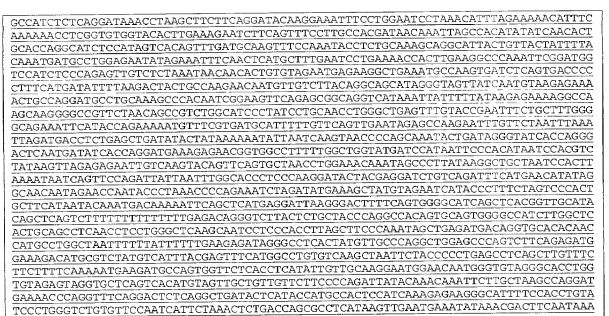
can be used in assay systems for functional analysis of various human disorders, which will help in understanding of pathology of the disease and development of new drug targets for various disorders.

NOV11

A disclosed NOV11 nucleic acid of 6540 nucleotides (also referred to as 87938450) encoding a novel transposase-like protein is shown in Table 11A. An open reading frame was identified beginning with an ATG initiation codon at nucleotides 758-760 and ending with a TGA codon at nucleotides 1175-1177. A putative untranslated region upstream from the initiation codon and downstream from the termination codon is underlined in Table 11A, and the start and stop codons are in bold letters.

Table 11A. NOV11 nucleotide sequence (SEQ ID NO:70).

CTGGAGTTCCTTTATTCTGGGGATAGCTCAAGTCCACTGCCAATGGCTGACAGTCATTAATACACAGGCAGAAAAAAGAA
ATAAGCTGCTGTGTCTGCAGTTGGGAGGGGAGCACTGGGAAGGACAGAATGGAAGTTACTGTATCCAGATACCAGCGGCC
TTTACATTTAAACATGGAGAGGAAGGAACAGGCAGATTAAAAAGTGAAAAATGGCAGTTTACAGAGAAGGCCTAACTGT
TGGAGAATGAGTACGAGATGAAGGGAAGCAGCTTTGATAGCAAACCAGGGGAATAAGGCAGTTATCTGCCAGTATCTACT
GCTTCAAAGAGAAGCTCAAGCATCATCTAAGTAGTTTTACACAGGGAGTGAGACTGAGTTTGGTGGGGATTTCATTGAGT
AATGGGATAAAAATTCAGGCACTGCTCATTCAGTTCCAAGGTTCTCTTGCAACCCAGTTTGAGCTGGAGGGAATTGTGT
TTTGGTACATATTTATGTTTGAATGCAAGCCAGCCCACATTCGACAGGCACGGAGCTCTTTCATGCTCAGAAAAGGGAAA
AAAAAGTTCCTGTTCTTGTATATTCTTTCATCCTAAACCTGAGACACTTAACAAGAAGCCGGTGTTGGCAAAGGTGTGTG
TGTGTGTGTGTGTCTGTGTGTGTGTGTCCTAACGAAATGCACATATTTGCTGCAGTGAAGGAGCCAGTTTTTCCATAAAT
GGCTAACAGGAATTTGATGAAGTGTTTGCAACATTAAATGTGTTGTGGGTCACGTTGTAACTTACATTGTTCCCCAGCCT
CCACTTTTCCTTGTTTCCTAACCAACCTCCATCCCGCCCACATGCCACATTCATCCAGGCCTTCAATGTTGTCTGTGTC
AGTTCCCATAAACTGGCTCAGGTTGTAGAAATGGTTAGTGAAGTCGGGCATCTCAGCCACTCCCCACCTCTTACTTCCCAA
GGTGTCTCATGTCACCAAATTACAAATCATCCACAAGCAGAAGATCAAATCAGGCTGACTAAAGCCATGTGGAATGTGG
ACACTTGGGGGCAGTTAAATACCTTACAGGTTTCTGCTGTAAGATTTGAAGCTTTGAAGGCAGAAATCAATGGCCAGATT
TTCAAAGGAAAGGTTACAGGTGTTGTCCAGGTGAGCCCCAGACAGATGGATCTGTGAAAGCAAGTGCCTGTGCAGGTGCA
GTGACTGCTCGGCCATATGTCCTGTACAGACATGGGCTGCAGAGGAAGGAACAAGACTGTGAGTCAAAGAAGACAGGCC
CGTGCAGCCATCCGTGCCTTACTTGTCTCCAGGTATATGGGGCAGATCTGTAAGTAGAGAATAAGAACAGCAGATGGGAT
TTTCCATGGGGACTCTACTTCCTACTCCAAGGCATTCAGAAACATGGCTAAAATGAAACCAGTGAATTTGGGGCCATAGA
GCTAATCTCAAAACCAAGAGAATGAAACTGCCAGGATGCATGAAGAGGGATGGCGAAGGCAGGCAGTAAGGAGGGGAAAC
TGAGTGGGCTCTGAATGTCACCTGCACGGTGTAGGCCCTCACGGCATCTTTCTGACCTCTAAATGTTGGAACACCCCAAC
AGGCCTGGGTCCTGCCTCCCCTGTCCCCTCTGCCACACTCTCTCTGGGTGAGCTCACTCAGCCCCACGCCTTTACATCCC
ATTTATGCACTGATGGCTCCTAACTCTAAATCTCCACCCCGACCCTTCTCCTGAGCTCCCGATTCAAAATCTTATGGCCT
GTTCATCCTCTTGGATATCTAATAGAGCTCCCAAAGTTAATGTGTCCAAACCTGAACCCCAGATTCGCCACTATGTTCCC
AAATCCCACTATGGGTAGTCTCCCCCATCTCAGAAAGTAACCCTCCATTTACCCAAGTGGTCTGGACAAAAGTTTGGGA
TTATCCTCAATTCTTTCTTTATCTCACATCCCGCATCTAATCCATCAGCAAGTTTCGTCAGCTCTCCCTGTAAAATGCA
TCCCATTCCTACTTCTTCATTGCTTCCACCACTACCAGCCCTGTTCAAAGCAACACCCTTTCTTTCCTTGATGACTGCAAT
GTTGTTGAGCTGACTGCCTTGATCCCATGCCTGCCACCTTGTTCTGTCTCCACACGGAAACTCAAGTGACTTTTTAAA
AGTATAAATAGATTAGCTGCCTGTTTCTTGCTCAAAACTTCTGCTGGTATTTCCTACTTTTAAAATGAAGTTCAAAGTCC
TAAAATAGCCTAACCTCTATTTACCACCCCCACCCCACCTCCTTCTATCTCCCTTTTGCCATTCCAGCCACACCAACCTC
CTGATCACCCTTCAAAATACATCACCTTGTTCCCTCTGTGGCATCTTGATATTTGTTGCTGTATCCACCTGGAAATCTTT
CACATTGCTCGTTCCCCTGATGCACTCAAAACTCTCTAATCCCACGTTCATCTTTGCAAAGAAGTCTTTCCTGACCACAG
ATTCTAAAGGAGACCAACCACCATCCAGCTCTTGGATCCTCCTCTTCTCTTCCCTTCTCCTGTTCCACGCATAGGGCACA
TTGATCATGGTTTTGGCTACCCAGTGTATTTAACATTCTTGTCCTATTTGAGAAAATTGAGACTCCCAAAGCAGAA
GGCAGTATAGTGAGTTTAATAGTGTTTCCCCTGATGTACATCTACCCAGAGCCTCAGAATATGACCTTAATTGGAAATAG
GTTCTTTGCAGCTATAATTAGTAAGGAGTGGAAGATGAAGTCATCCTGAATTTAGGGTGGGCCCTAATTCCAATGACTG
GCATCCTTATGAGATAATGGAGAAGGAGATTTGGACACAGACATGAAGACATGCAGGAAAGAAGGCCACCTAGTAATGGA
GGCAGGGTGACTCATGGAGCCACAAGCCAACGGACATCAAGTACCACTGGCCCCCATCAAAAACTTTAAAAAGCAGGGA
AAGGTTCTTCTCTAGAGCCTCCAGAGGGAACAGGACTCTGTTAACACCTCAATCTCAGCCTTCCAGCCTCCAGACTGTGA
GAGAATAAAGCCATCAAGTTTGTGGTTATTAGTTACAGCAGGCTTAGGAAACTAATACAGCCAAACATTTCTCTAGATGC
TCAGTAACCAGGGCACAAGACAGAGACCCACACCCCCAGTCAGATGATTCTGCATGAGACTTCCATTGTAGATCTGAGT
GCATTGAGGAGCTCACCCCAGCAGTTCCTATCATCCCAGCTCAGGCTCAGACATCAAGAAGCAGGAGACAAGCCATCT
CTGTTGTGTCCTGTCAAAACCCTGAGCCATAGACTTCATGGGCATAACAAAATGGTTTGTGTTGAGCCCATAAAGTTGG
AGTGCTTTGTTGTACAGCAATAGTAACTGCAACAAAAATCAAAATAATTCCTCTCTGATGGTGGGGCATGGGGAAGATGA
AGGAAAGAGATATAGTGAATCACATCTTTGTCAGAAAGACAGTGGGTTCATTTGAGTAGTTGGATTATGTATTTCCCAGA



In a search of public sequence databases, the NOV11 nucleic acid sequence has no hits using an Expect value of 1.0. Public nucleotide databases include all GenBank databases and the GeneSeq patent database.

A disclosed NOV11 polypeptide (SEQ ID NO:71) encoded by SEQ ID NO:70 has 139 amino acid residues and is presented in Table 11B using the one-letter amino acid code. SignalP, Psort and/or Hydropathy results predict that NOV11 has no known signal peptide and is likely to be localized to the mitochondrial matrix space with a certainty of 0.4344. In alternative embodiments, the NOV11 protein is localized to a microbody (peroxisome) with a certainty of 0.3191; a lysosome (lumen) with a certainty of 0.1589; or the mitochondrial inner membrane with a certainty of 0.1162. NOV11 has a molecular weight of 15546.1 Daltons.

Table 11B. Encoded NOV11 protein sequence (SEQ ID NO:71).

MCCGSRCNLHCSPASTFPCFLTNLHPAPHATFIQAFNRSAVSSHKLAQVVEMVSEVGHLSHSHLLLPKVSHVTKLQIIH
KQKIKSRLTKAMWNVDTWGQLNTLQVSAVRFEALKAEINGQIFKGKGYRCVQVSPRQMDL

PROSITE analysis of NOV11 predicts that the NOV11 protein has one N-glycosylation site (Pattern-ID: ASN_GLYCOSYLATION PS00001 (Interpro)); two Protein kinase C phosphorylation sites (Pattern-ID: PKC_PHOSPHO_SITE PS00005 (Interpro)); and two N-myristoylation sites (Pattern-ID: MYRISTYL PS00008 (Interpro)),

Table 11C lists the domain description from DOMAIN analysis results against NOV11. This indicates that the NOV11 sequence has properties similar to those of other proteins known to contain this transposase_17 domain.

Table 11C. Domain Analysis of NOV11

PFAM HMM Domain Analysis of NOV11

Model	Description	Score	E-value
Transposase_17 (InterPro)	Transposase IS200 like	-42.6	9.5

PRODOM Domain Analysis of NOV11

Sequences producing High-scoring Segment Pairs:	High Score	Smallest Sum Probability P(N)
prdm:29481 p36 (1) AADR_RHOPA - ANAEROBIC AROMATIC DEGRA...	51	0.61
prdm:20370 p36 (1) YVAU_VACCC - HYPOTHETICAL 8.8 KD PROT...	49	0.80
prdm:44828 p36 (1) YM91_SCHPO - HYPOTHETICAL 91 KD PROTE...	49	0.80
prdm:28458 p36 (1) PR1_MEDTR - PATHOGENESIS-RELATED PROT...	47	0.93
prdm:29156 p36 (1) POL_SMRVH - POL POLYPROTEIN (REVERSE ...	46	0.97

BLOCKS Protein Domain Analysis

AC#	Description	Strength	Score
BL01280E	0 Glucose inhibited division protein A family p	1592	1031
BL00884D	0 Osteopontin proteins.	1466	1027
BL00130E	0 Uracil-DNA glycosylase proteins.	1320	1006
BL00441E	0 Chalcone and stilbene synthases proteins.	2040	1000

Table 11D provides percent homology to the domains identified in Table 11C.

Table 11D. ProDom BLASTP results for NOV11

ProDom Identifier	Protein/ Organism	Length (aa)	Identity (%)	Positive (%)	Expect
prdm:29481	p36 (1) AADR_RHOPA - DNA-binding ANAEROBIC AROMATIC DEGRADATION REGULATOR	47	12/31 (38%)	13/31 (41%)	0.95
prdm:20370	p36 (1) YVAU_VACCC HYPOTHETICAL 8.8 KD PROTEIN	53	10/20 (50%)	12/20 (60%)	1.6
prdm:44828	p36 (1) YM91_SCHPO - HYPOTHETICAL 91 KD PROTEIN IN COB INTRON, HYPOTHETICAL PROTEIN; MITOCHONDRION	34	8/20 (40%)	13/20 (65%)	1.6
prdm:28458	p36 (1) PR1_MEDTR - PATHOGENESIS-RELATED PROTEIN PR-1 PRECURSOR	45	12/26 (46%)	16/26 (61%)	2.7
prdm:29156	p36 (1) POL_SMRVH - POL POLYPROTEIN (REVERSE TRANSCRIPTASE (EC 2.7.7.49); ENDONUCLEASE)	34	9/28 (32%)	14/28 (50%)	3.5

NOV11 polypeptide sequence produced no hits in a BLASTP search for homology (Expect value setting = 1.0) to the GenBank and EMBL public databases. Other BLAST results did find homologous sequences from the Patp database, which is a proprietary database that contains sequences published in patents and patent publications. According to a BlastP analysis, NOV11 has 38 of 64 aa residues(59%) identical to, and 49 of 64 (76%) positive with, the 102 aa Human protein sequence SEQ ID NO:18455 from PN=EP1074617-A2 (patp: AAB95670, Expect = 4.6e-16); 35 of 58 aa residues(60%) identical to, and 42 of 58 (72%) positive with, the 101 aa an secreted protein, SEQ ID NO: 4718 from PN=EP1033401-A2 (patp: AAG00637, Expect = 5.1e-15);20 of 61 aa residues(32%) identical to, and 35 of 61 (57%) positive with, the 136 aa Arabidopsis thaliana protein fragment SEQ ID NO: 42276 (patp:AAG34708, Expect = 0.51); 20 of 61 (32%) identical to, and 35 of 61 (57%) positive with, the 150 aa Arabidopsis thaliana protein fragment SEQ ID NO: 42275 (patp:AAG34707, Expect = 0.71); 20 of 61 (32%) identical to, and 35 of 61 (57%) positive with, the 162 aa Arabidopsis thaliana protein fragment SEQ ID NO: 42274 (patp:AAG34706, Expect = 0.89); 20 of 61 (32%) identical to, and 35 of 61 (57%) positive with, the 270 aa Arabidopsis thaliana protein fragment SEQ ID NO: 21878 (patp:AAG19901, Expect = 2.5); 13 of 36 (36%) identical to, and 17 of 36 (47%) positive with, the 66 aa Human endometrium tumour EST encoded protein 343 (patp:AAY60283, Expect = 4.3); 10 of 26 (38%) identical to, and 18 of 26 (69%) positive with, the 64 aa Gene 8 human secreted protein homologous amino acid sequence #113 - Bos taurus (patp:AAB39364, Expect = 5.6); and 10 of 26 (38%) identical to, and 18 of 26 (69%) positive with, the 64 aa Human secreted protein sequence encoded by gene 8 SEQ ID NO:114 (patp:AAB39365, Expect = 5.6). Patp results include those listed in Table 11E.

Table 11E. Patp alignments of NOV11

Sequences producing High-scoring Segment Pairs:	High Score	Smallest Sum Prob. P(N)
patp:AAG34708 Arabidopsis thaliana protein fragment SEQ I...	74	0.40
patp:AAG34707 Arabidopsis thaliana protein fragment SEQ I...	74	0.51
patp:AAG34706 Arabidopsis thaliana protein fragment SEQ I...	74	0.59
patp:AAG19901 Arabidopsis thaliana protein fragment SEQ I...	74	0.91
patp:AAY60283 Human endometrium tumour EST encoded protei...	53	0.99

The disclosed NOV11 nucleic acid encoding a transposase-like protein includes the nucleic acid whose sequence is provided in Table 11A, or a fragment thereof. The invention also includes a mutant or variant nucleic acid any of whose bases may be changed from the corresponding base shown in Table 11A while still encoding a protein that maintains its transposase-like activities and physiological functions, or a fragment of such a nucleic acid. The

invention further includes nucleic acids whose sequences are complementary to those just described, including nucleic acid fragments that are complementary to any of the nucleic acids just described. The invention additionally includes nucleic acids or nucleic acid fragments, or complements thereto, whose structures include chemical modifications. Such modifications include, by way of nonlimiting example, modified bases, and nucleic acids whose sugar phosphate backbones are modified or derivatized. These modifications are carried out at least in part to enhance the chemical stability of the modified nucleic acid, such that they may be used, for example, as antisense binding nucleic acids in therapeutic applications in a subject. In the mutant or variant nucleic acids, and their complements, up to about 60 % percent of the bases may be so changed.

The disclosed NOV11 protein of the invention includes the transposase-like protein whose sequence is provided in Table 11B. The invention also includes a mutant or variant protein any of whose residues may be changed from the corresponding residue shown in Table 11B while still encoding a protein that maintains its transposase-like activities and physiological functions, or a functional fragment thereof. In the mutant or variant protein, up to about 60 % percent of the residues may be so changed.

The invention further encompasses antibodies and antibody fragments, such as F_{ab} or $(F_{ab})_2$, that bind immunospecifically to any of the proteins of the invention.

The above defined information for this invention suggests that this transposase-like protein (NOV11) may function as a member of a "transposase family". Therefore, the NOV11 nucleic acids and proteins identified here may be useful in potential therapeutic applications implicated in (but not limited to) various pathologies and disorders as indicated below. The potential therapeutic applications for this invention include, but are not limited to: transposase related research tools, for all tissues and cell types composing (but not limited to) those defined herein.

The protein similarity information, expression pattern, cellular localization, and map location for the protein and nucleic acid disclosed herein suggest that this novel intracellular transposase domain containing protein-like NOV11 protein may have important structural and/or physiological functions characteristic of the novel transposase domain containing protein family. Therefore, the NOV11 nucleic acids and proteins are useful in potential diagnostic and therapeutic applications and as a research tool. These include serving as a specific or selective nucleic acid or protein diagnostic and/or prognostic marker, wherein the presence or amount of the nucleic acid or the protein are to be assessed. These also include potential therapeutic applications such as the following: (i) a protein therapeutic, (ii) a small molecule drug target, (iii) an antibody target (therapeutic, diagnostic, drug targeting/cytotoxic antibody), (iv) a nucleic acid

useful in gene therapy (gene delivery/gene ablation). (v) an agent promoting tissue regeneration *in vitro* and *in vivo*, and (vi) a biological defense weapon.

The NOV11 nucleic acids and proteins of the invention are useful in potential therapeutic applications including but not limited to those provided in Example 2, and/or other pathologies and disorders. For example, a cDNA encoding the transposase-like protein (NOV11) may be useful in gene and protein therapy, and the transposase-like protein (NOV11) may be useful when administered to a subject in need thereof. The NOV11 nucleic acid encoding the transposase-like protein, and the transposase-like protein of the invention, or fragments thereof, may further be useful in diagnostic applications, wherein the presence or amount of the nucleic acid or the protein are to be assessed.

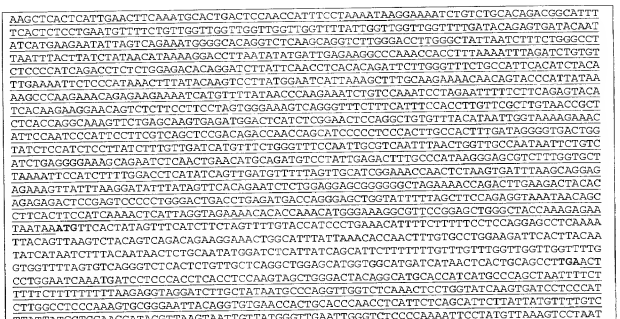
NOV11 nucleic acids and polypeptides are further useful in the generation of antibodies that bind immuno-specifically to the novel NOV11 substances for use in therapeutic or diagnostic methods. These antibodies may be generated according to methods known in the art, using prediction from hydrophobicity charts, as described in the "Anti-NOVX Antibodies" section below. The disclosed NOV11 protein has multiple hydrophilic regions, each of which can be used as an immunogen. In one embodiment, a contemplated NOV11 epitope is from about amino acids 25 to 45. In additional embodiments, NOV11 epitopes are from about amino acids 70 to 105 and from about amino acids 11- to 139. These novel proteins can be used in assay systems for functional analysis of various human disorders, which will help in understanding of pathology of the disease and development of new drug targets for various disorders.

NOV12

A disclosed NOV12 nucleic acid of 2760 nucleotides (also referred to as 87917235 or 13373979) encoding a novel Novel Leucine Zipper Containing Type II membrane like protein-like protein is shown in Table 12A. An open reading frame was identified beginning with an ATG initiation codon at nucleotides 1789-1791 and ending with a TGA codon at nucleotides 2101-2103. A putative untranslated region upstream from the initiation codon and downstream from the termination codon is underlined in Table 12A, and the start and stop codons are in bold letters.

Table 12A. NOV12 Nucleotide Sequence (SEQ ID NO:72)

TCTGCCTCCTGGGTTGAAGCGATTCTTCTGCCTCAGCCTCCTGAGTAGCTGGGACTACAGCAGGCGCCACTACGCCTGGC
TAATTTTTGTATTTTTAGTAGAGACAGGGTTTCACCATATTGGCCAGGATGGTTTCAAACTCCTGACCTCATGATCTGCCC
ACCTAGGCCTCCCAAAGTGCTGGGATTACAGGCGTGAGCCACCGCGCCTGGTGAGGACTCCATTTTCTACCCCTAGGCTAA
AGAGCCTGGAGGATTATAGCTTACAGAGCAGAGAAGAACTCTGATACTCATACCTGCATAGTGCTAGCTAGTCAGTAGACA
ATACTTAGATAATTCATTTTCTGATTTCTGACATTAGTGAGAGGTTGGGGTTTTGTTTGTTTAATAACAGCCTTCATTTAG
ATCTTTGCAAACAGCCTTGAATGAGGAATGTCCTTATGTTTCAGGGAACATATCAGGCCTGGAAGCAGCTTTTTTAGGATA



The NOV12 nucleic acid was identified on chromosome 17 as run against the Genomic Daily Files made available by GenBank or from files downloaded from the individual sequencing centers. Exons were predicted by homology and the intron/exon boundaries were determined using standard genetic rules. Exons were further selected and refined by means of similarity determination using multiple BLAST (for example, tBlastN, BlastX, and BlastN) searches, and, in some instances. GeneScan and Grail. Expressed sequences from both public and proprietary databases were also added when available to further define and complete the gene sequence. The DNA sequence was then manually corrected for apparent inconsistencies thereby obtaining the sequences encoding the full-length protein. The NOV12 nucleic acid was further mapped to the p11 region of chromosome 17, a locus associated with prostate cancer (OMIM 176807) and congenital slow-channel myosthenic syndrome (OMIM 601462).

A disclosed NOV12 polypeptide (SEQ ID NO:73) encoded by SEQ ID NO:72 is 104 amino acid residues and is presented using the one-letter amino acid code in Table 12B. SignalP, Psort and/or Hydropathy results predict that NOV12 does not contain a known signal peptide and in the likely to be localized in the cytoplasm with a certainty of 0.8387 predicted by PSORT. In alternative embodiments, NOV12 is likely to be localized to the mitochondrial inner membrane with a certainty of 0.8387, to a microbody (peroxisome) with a certainty of 0.7480, the plasma membrane with a certainty of 0.4400, or the mitochondrial intermembrane space with

a certainty of 0.3751. The NOV12 hydropathy profile is characteristic of the 'leucine zipper' gene family. A NOV12 polypeptide has a molecular weight of 11855.7 Daltons.

Table 12B. Encoded NOV12 protein sequence (SEQ ID NO:73).

```
MFTIVSSSSFVPSLKHFLFPPGASKLQLSLQSDRRKLAFIKHQLCAWKIHLQYHNLYNNSAIWISLSAFFFCLFGWLVLV
VLVSGSHSVAQAGAWWHDHNSLQP
```

In a search of sequence databases, it was found, for example, that the nucleic acid sequence of this invention has 168 of 252 bases (66%) identical to a gb:GENBANK-ID:HS435C23|acc:Z92844.1 mRNA from Homo sapiens (Human DNA sequence from PAC 435C23 on chromosome X. Contains ESTs). No sequences were found in the EMBL, PIR or GenBank databases that had homology to the NOV12 polypeptide in an unfiltered BLASTP search (expectation value=1.0 for input parameter).

Table 12C lists the domain description from DOMAIN analysis results against NOV12. This indicates that the NOV12 sequence has properties similar to those of other proteins known to contain this domain.

Table 12C. Domain Analysis of NOV12

```
PRODOM Analysis
prdm:49789 p36 (1) RED1_HUMAN // DOUBLE-STRANDED RNA-SPECIFIC EDITASE 1 (DSRNA
ADENOSINE DEAMINASE) (RNA EDITING ENZYME 1). RNA EDITING; HYDROLASE; ZINC; RNA-
BINDING; REPEAT; ALTERNATIVE SPLICING, 55 aa.
Expect = 0.012, Identities = 12/23 (52%), Positives = 15/23 (65%)
 for aa of Query: 82 to 104, Sbjct: 1 to 23

prdm:5031 p36 (5) NU4M(5) // OXIDOREDUCTASE NADH-UBIQUINONE CHAIN NAD UBIQUINONE
MITOCHONDRION, 43 aa.
Expect = 0.63, Identities = 9/22 (40%), Positives = 12/22 (54%)
for aa of Query: 56 to 77, Sbjct: 20 to 41

prdm:22836 p36 (1) NU1C_SYNY3 // NADH-PLASTOQUINONE OXIDOREDUCTASE CHAIN 1 (EC
1.6.5.3). OXIDOREDUCTASE; NAD; PLASTOQUINONE; TRANSMEMBRANE, 28 aa.
Expect = 0.83, Identities = 10/19 (52%), Positives = 14/19 (73%)
for aa of Query: 8 to 26, Sbjct: 9 to 27

PROSITE Analysis
Pattern Name                                    Pattern        Position of NOV12
ASN_GLYCOSYLATION PS00001 (Interpro) PDOC00001  N[^P][ST][^P]  58
PKC_PHOSPHO_SITE PS00005 (Interpro) PDOC00005   [ST].[RK]      13, 32
LEUCINE_ZIPPER PS00029 (Interpro) PDOC00029  L.{6}L.{6}L.{6}L   30

BLOCKS Analysis
AC#          Description                                Strength  Score
BL00435D  Peroxidases proximal heme-ligand proteins.      1230    1101
BL00604C  Synaptophysin / synaptoporin proteins.          1917    1030
BL00439D  Acyltransferases ChoActase / COT / CPT family   1332    1029
BL00177C  DNA topoisomerase II proteins.                  1219    1021
BL00487H  IMP dehydrogenase / GMP reductase proteins.     1405    1016

PFam Analysis
[no hits above thresholds]
```

Patp BLAST results for NOV12 include those listed in Table 12D.

Table 12D. Patp alignments of NOV12

Sequences producing High-scoring Segment Pairs:	Score	Smallest Sum Prob.
patp:AAG03340 Human secreted protein, SEQ ID NO: 7421 - H. .	68	0.00028
patp:AAY27571 Human secreted protein encoded by gene No. ...	92	0 00071
patp:AAB95648 Human protein sequence SEQ ID NO:18400 - Ho...	85	0 0010
patp:AAB42720 Human ORFX ORF2484 polypeptide sequence SEQ...	81	0.0023
patp:AAG00591 Human secreted protein, SEQ ID NO: 4672 - H...	81	0.0023

A structure, referred to as the "leucine zipper", has been proposed to explain how some eukaryotic gene regulatory proteins work. The leucine zipper consist of a periodic repetition of leucine residues at every seventh position over a distance covering eight helical turns. The segments containing these periodic arrays of leucine residues seem to exist in an alpha-helical conformation. The leucine side chains extending from one alpha-helix interact with those from a similar alpha helix of a second polypeptide, facilitating dimerization; the structure formed by cooperation of these two regions forms a coiled coil.

The leucine zipper pattern is present in many gene regulatory proteins, *e.g.* the CCATT-box and enhancer binding protein (C/EBP), the cAMP response element (CRE) binding proteins (*e.g.* CREB, CRE-BP1, ATFs), the Jun/AP1 family of transcription factors, the yeast general control protein GCN4, the fos oncogene and the fos-related proteins fra-1 and fos B. the C-myc, L-myc and N-myc oncogenes, and the octamer-binding transcription factor 2 (Oct-2/OTF-2). Thus, leucine zipper-like proteins are involved in cell proliferation, migration and differentiation. Leucine zipper-like proteins may thus be implicated in the onset and/or maintenance of diseases including cancer, *e.g.* prostate cancer. diabetes, abnormal wound healing, congenital slow-channel myosthenic syndrome, inflammation and/or other diseases and disorders. The consensus pattern for leucine zipper-like proteins is: L-x(6)-L-x(6)-L-x(6)-L.

The above defined information for this invention suggests that these Leucine Zipper Containing Type II membrane protein-like proteins (NOV12) may function as a member of a "leucine zipper family". Therefore, the NOV12 nucleic acids and proteins identified here may be useful in potential therapeutic applications implicated in (but not limited to) various pathologies and disorders as indicated herein. The potential therapeutic applications for this invention include, but are not limited to: cancer. *e.g.* prostate cancer, diabetes, abnormal wound healing, congenital slow-channel myosthenic syndrome, inflammation and/or other diseases and disorders.

The novel nucleic acid encoding a Leucine Zipper Containing Type II membrane like protein-like NOV12 protein includes the nucleic acid whose sequence is provided in Table 12A, or a fragment thereof. The invention also includes a mutant or variant nucleic acid any of whose

bases may be changed from the corresponding base shown in Table 12A while still encoding a protein that maintains its Leucine Zipper Containing Type II membrane like protein-like activities and physiological functions, or a fragment of such a nucleic acid. The invention further includes nucleic acids whose sequences are complementary to the Leucine Zipper Containing Type II membrane like protein-like NOV12 nucleic acid sequence, including nucleic acid fragments that are complementary to any of the nucleic acids just described. The invention additionally includes nucleic acids or nucleic acid fragments, or complements thereto, whose structures include chemical modifications. In the mutant or variant NOV12 nucleic acids, and their complements, up to about 34% of the bases may be so changed.

The novel protein of the invention includes the Leucine Zipper Containing Type II membrane like protein-like NOV12 protein whose sequence is provided in Table 12B. The invention also includes a mutant or variant protein any of whose residues may be changed from the corresponding NOV12 residue while still encoding a protein that maintains its Leucine Zipper Containing Type II membrane like protein-like activities and physiological functions, or a functional fragment thereof. In the mutant or variant protein, up to about 37% of the NOV12 amino acid residues may be so changed.

The NOV12 nucleic acids and proteins of the invention are useful in potential therapeutic applications implicated in cancer, *e.g.* prostate cancer, diabetes, abnormal wound healing, congenital slow-channel myosthenic syndrome, inflammation and/or other pathologies and disorders. For example, a cDNA encoding the leucine zipper-like NOV12 protein may be useful in detecting prostate cancer, and the leucine zipper-like protein may be useful when administered to a subject in need thereof. By way of nonlimiting example, the compositions of the present invention will have efficacy for treatment of patients suffering from prostate cancer or congenital slow-channel myosthenic syndrome. The NOV12 nucleic acid encoding leucine zipper-like protein, and the leucine zipper-like protein of the invention, or fragments thereof, may further be useful in diagnostic applications, wherein the presence or amount of the nucleic acid or the protein are to be assessed. Additional disease indications and tissue expression for NOV12 is presented in Example 2.

NOV12 nucleic acids and polypeptides are further useful in the generation of antibodies that bind immuno-specifically to the novel NOV12 substances for use in therapeutic or diagnostic methods. These antibodies may be generated according to methods known in the art, using prediction from hydrophobicity charts, as described in the "Anti-NOVX Antibodies" section below. For example the disclosed NOV12 proteins have multiple hydrophilic regions, each of which can be used as an immunogen. In one embodiment, a contemplated NOV12 epitope is from about amino acids 20 to 40. In additional embodiments, NOV12 epitopes are

from about amino acids 20 to 25 and from about amino acids 30 to 40. This novel protein also has value in development of powerful assay systems for functional analysis of various human disorders, which will help in understanding of pathology of the disease and development of new drug targets for various disorders.

NOV13

A disclosed NOV13 nucleic acid of 1183 nucleotides (also referred to as 87919652) encoding a novel tyrosine kinase-like protein is shown in Table 13A. An open reading frame was identified beginning with an ATG initiation codon at nucleotides 398-400 and ending with a TAG codon at nucleotides 1181-1183. A putative untranslated region upstream from the initiation codon is underlined in Table 13A, and the start and stop codons are in bold letters.

Table 13A. NOV13 nucleotide sequence (SEQ ID NO:74).

AGCTAGAGCTCCAAGGACCCCACGCCTGTGTCTCTGTGACAGAGCTCAAAGGGCCCTGGGCCTTCCCTCCCTGGCTCGGC
TGTGCTTGGGAGGGTTCCCCAGTCCAGAATCCCTAAGGAGCATGGGGCAGCTGATCCATCCCTGGTGTACAAACTGCTGA
CTGCAGACAGATGCTGAGCTACCCAAACCAACACCTAGCCTCTCCCTGAAGATCCTCCCAGGCTGAGAGAGTTCTGGGTG
TCCTAGGACCAAGGACACTGGCAGACTTCCAGAAGGGCCCCCAAAGCCCTAACCTGTCCAGCCAGAGCATGCGTCTCAGC
AGAGCTGTCTTCCCAAGCCTTTGATGACAAACCAATTTCCCTCGATGATGTGCTTCTGAGTGCTCTGCTGAGGAACAATG
GGAAGTCTGCCCAGCAGAAGAAAATCTCTGCCAAGCCCAAGCTTGAGTTCCTCTGTCCAAGGCCAGGGACCTGTGACCAT
GGAAGCAGAGAGAAGCAAGGCCACAGCCGTCGCCCTGGGCAGTTCCCGGCAGGTGGCCCGGCCGAGCTGTCGCTGAGAC
TCGGGAGCCATTGACCATCGTCTCTGAGGATGGAACTGGTGACGGTGCTGTCTGAAGTCTCAGGCAGAGAGTATAAC
ATCCCCAGCGTCCACGTGGCAAAGTCTCCCATGGGTGGCTGTATGAGGGCCTGAGCAGGGAGAAAGCAGAGGAACTGCT
GTTGTTACCTGGGAACCCTGGAGGGGCCTTCCTCATCCGGGAGAGCCAGACCAGGAGAGGCTCTTACTCTCTGTCAGTCC
GCCTCAGCCGCCCTGCATCCTGGGACCGGATCAGACACTACAGGATCCACTGCCTTGACAATGGCTGGCTGTACATCTCA
CCGCGCCTCACCTTCCCCTCACTCCAGGCCCTGGTGGACCATTACTCTGAGCTGGCGGATGACATCTGCTGCCTACTCAA
GGAGCCCTGTGTCCTGCAGAGGGCTGGCCCGCTCCCTGGCAAGGATATACCCCTACCTGTGACTGTGCAGAGGACACCAC
TCAACTGGAAAGAGCTGGACAGCTCCCTCCTGTTTTCTGAAGCTGCCACAGGGGAGGAGTCTCTTCTCAGTGAGGGTCTC
CGGGAGTCCCTCAGCTTCTACATCAGCCTGAATGACGAGGCTGTCTCTTTGGATGATGCCTAG

The NOV13 nucleic acid was identified on chromosome 20 by comparing it to the human genome database. Exons were predicted by homology and the intron/exon boundaries were determined using standard genetic rules. Exons were further selected and refined by means of similarity determination using multiple BLAST (for example, tBlastN, BlastX, and BlastN) searches, and, in some instances, GeneScan and Grail. Expressed sequences from both public and proprietary databases were also added when available to further define and complete the gene sequence. The DNA sequence was then manually corrected for apparent inconsistencies thereby obtaining the sequences encoding the full-length protein.

A disclosed NOV13 polypeptide (SEQ ID NO:75) encoded by SEQ ID NO:74 has 261 amino acid residues and is presented in Table 13B using the one-letter amino acid code. SignalP, Psort and/or Hydropathy results predict that NOV13 does not have a known signal peptide and is likely to be localized in the mitochondrial matrix space with a certainty of 0.4737.

In an alternative embodiment, NOV13 is likely to be localized in the cytoplasm with a certainty of 0.4500.

Table 13B. Encoded NOV13 protein sequence (SEQ ID NO:75).

```
MGSLPSRRKSLPSPSLSSSVQGQGPVTMEAERSKATAVALGSFPAGGPAELSLRLGEPLTIVSEDGDWWTVLSEVSGREYN
IPSVHVGKVSHGWLYEGLSREKAEELLLLPGNPGGAFLIRESQTRRGSYSLSVRLSRPASWDRIRHYRIHCLDNGWLYISP
RLTFPSLQALVDHYSELADDICCLLKEPCVLQRAGPLPGKDIPLPVTVQRTPLNWKELDSSLLFSEAATGEESLLSEGLRE
SLSFYISLNDEAVSLDDA
```

The reverse complement for NOV13 is presented in Table 13C.

Table 13C. NOV13 reverse complement (SEQ ID NO:76).

```
CTAGGCATCATCCAAAGAGACAGCCTCGTCATTCAGGCTGATGTAGAAGCTGAGGGACTCCCGGAGACCCTCACTGAGAA
GAGACTCCTCCCCTGTGGCAGCTTCAGAAAACAGGAGGGAGCTGTCCAGCTCTTTCCAGTTGAGTGGTGTCCTCTGCACA
GTCACAGGTAGGGGTATATCCTTGCCAGGGAGCGGGCCAGCCCTCTGCAGGACACAGGGCTCCTTGAGTAGGCAGCAGAT
GTCATCCGCCAGCTCAGAGTAATGGTCCACCAGGGCCTGGAGTGAGGGAAGGTGAGGCGCGGTGAGATGTACAGCCAGC
CATTGTCAAGGCAGTGGATCCTGTAGTGTCTGATCCGGTCCCAGGATGCAGGGCGGCTGAGGCGGACTGACAGAGAGTAA
GAGCCTCTCCTGGTCTGGCTCTCCCGGATGAGGAAGGCCCCTCCAGGGTTCCCAGGTAACAACAGCAGTTCCTCTGCTTT
CTCCCTGCTCAGGCCCTCATACAGCCACCCATGGGAGACTTTGCCCACGTGGACGCTGGGGATGTTATACTCTCTGCCTG
AGACTTCAGACAGCACCGTCCACCAGTCTCCATCCTCAGAGACGATGGTCAATGGCTCCCCGAGTCTCAGCGACAGCTCG
GCCGGGCCACCTGCCGGGAAACTGCCCAGGGCCACGGCTGTGGCCTTGCTTCTCTCTGCTTCCATGGTCACAGGTCCCTG
GCCTTGGACAGAGGAACTCAAGCTTGGGCTTGGCAGAGATTTTCTTCTGCTGGGCAGACTTCCCATTGTTCCTCAGCAGA
GCACTCAGAAGCACATCATCGAGGGAAATTGGTTTGTCATCAAAGGCTTGGGAAGACAGCTCTGCTGAGACGCATGCTCT
GGCTGGACAGGTTAGGGCTTTGGGGGCCCTTCTGGAAGTCTGCCAGTGTCCTTGGTCCTAGGACACCCAGAACTCTCTCA
GCCTGGGAGGATCTTCAGGGAGAGGCTAGGTGTTGGTTTGGGTAGCTCAGCATCTGTCTGCAGTCAGCAGTTTGTACACC
AGGGATGGATCAGCTGCCCCATGCTCCTTAGGGATTCTGGACTGGGGAACCCTCCCAAGCACAGCCGAGCCAGGGAGGGA
AGGCCCAGGGCCCTTTGAGCTCTGTCACAGAGACACAGGCGTGGGGTCCTTGGAGCTCTAGCT
```

In a search of public sequence databases, the NOV13 amino acid sequence has 175 of 197 amino acid residues (89%) identical to, and 175 residues (89 %) positive with, the 197 amino acid residue human protein tyrosine kinase (Accession No. Q9H135). Public amino acid databases include the GenBank databases, SwissProt, PDB and PIR.

It was also found that NOV13 had homology to the amino acid sequences shown in the BLASTP data listed in Table 13D.

Table 13D. BLAST results for NOV13

Gene Index/ Identifier	Protein/ Organism	Length (aa)	Identity (%)	Positives (%)	Expect
Q9H6Q3; AK025645; BAB15201.1	CDNA: FLJ21992 FIS, CLONE HEP06554. homo sapiens. 6/2001	261	260/261 (100%)	260/261, (100%)	1e-149
Q9H135; AL050318; CAB75365.1	DJ977B1.1 (NOVEL PROTEIN TYROSINE KINASE WITH SRC HOMOLOGY DOMAIN 2DOMAINS) (FRAGMENT). homo sapiens. 6/2001	197	196/197 (99%)	196/197, (99%)	1e-113

Q9D1Z9; AK020837; BAB32223 1	A930009E21RIK PROTEIN. mus musculus. 6/2001	179	148/181 (82%)	159/181, (88%)	8e-79
Q60898; U29056; AAA82756.1	SRC-LIKE ADAPTER PROTEIN. mus musculus. 6/2001	281	106/253 (42%)	148/253, (58%)	2e-47
Q13239; U30473; AAC50357.1; AAC27662.1; BAA13758.1	PUTATIVE SRC-LIKE ADAPTER PROTEIN (SLAP). homo sapiens. 6/2001	276	96/219 (44%)	135/219, (62%)	1e-46

The homology of these sequences listed in Table 13D is shown graphically in the ClustalW analysis shown in Table 13E.

Table 13E Information for the ClustalW proteins

```
1)  NOV13 (SEQ ID NO:75)
2)  Q9H6Q3 (SEQ ID NO:77)
3)  Q9H135 (SEQ ID NO:78)
4)  Q9D1Z9 (SEQ ID NO:79)
5)  Q60898 (SEQ ID NO:80)
6)  Q13239 (SEQ ID NO:81)

NOV13            1   MGSLPSRRKSLPSPSLSSSVQGQGPVTMEAERSKATAVALGSFPAGGPAELSLRLGEPLT  60
Q9H6Q3           1   MGSLPSRRKSLPSPSLSSSVQGQGPVTMEAERSKATAVALGSFPAGGPAELSLRLGEPLT  60
Q9H135           1   ------------------------------------------------------------  1
Q9D1Z9           1   ------------------------------------------------------------  1
Q60898           1   MG---NSMKSTSPPS-------ERPLSSSEGLESDFLAVLTDYPSPDISPPIFRRGEKLR  50
Q13239           1   MG---NSMKSTPAPA-------ERPLPNPEGLDSDFLAVLSDYPSPDISPPIFRRGEKLR  50

NOV13           61   IVSEDGDWWTVLSEVSGREYNIESVHVAKVSHGWLVEGLSRGKAEELLLGPNPGSAFLI  120
Q9H6Q3          61   IVSEDGDWWTVLSEVSGREYNIESVHVAKVSHGWLVEGLSRGKAEELLLGPNPGSAFLI  120
Q9H135           1   ----DGDWWTVLSEVSGREYNIESVHVAKVSHGWLVEGLSRGKAEELLLGPNPGSAFLI   56
Q9D1Z9           1   ------------------MESVYVAKVAHGWLVEGLSRGKAEELLLGPNPGSAFLI   39
Q60898          51   VISDEGGWWKAISLSTGRESYIPGICVARVEHGWLVEGLSRGKAEELLGPDTKIGSFMI  110
Q13239          51   VISDEGGWWKAISLSTGRESYIPGICVARVEHGWLVEGLSRGKAEELLGPDTKVGSFMI  110

NOV13           121  RESQTRRGGYSLSVRLSRPASWDRIRHYRIHCLDNGWLYISPRLTFPSLQALVDHYSEMA  180
Q9H6Q3          121  RESQTRRGGYSLSVRLSRPASWDRIRHYRIHCLDNGWLYISPRLTFPSLQALVDHYSEMA  180
Q9H135           57  RESQTRRGGYSLSVRLSRPASWDRIRHYRIHCLDNGWLYISPRLTFPSLQALVDHYSEMA  116
Q9D1Z9           40  RESQTRRGGYSLSVRLSRPASWDRIRHYRIQRLDNGWLYISPRLTFPSLHALVDHYSEMA  99
Q60898          111  RESETKKGGYSLSVR-PR------QVKHYRIFRLPNGWYISPRLTFQCLEDLVHYSEVA  164
Q13239          111  RESETKKGGYSLSVR-PR------QVKHYRIFRLPNGWYISPRLTFQCLEDLVNYSEVA  164

NOV13           181  DDICCLLKEPCVLQRAGPLPGKDIP-----LDVTVQRTPLNKRELDSSLLFSPA-TTG--  232
Q9H6Q3          181  DDICCLLKEPCVLQRAGPLPGKDIP-----LDVTVQRTPLNKRELDSSLLFSPA-TTG--  232
Q9H135          117  DDICCLLKEPCVLQRAGPLPGKDIP-----LDVTVQRTPLNKRELDSSLLFSPA-TTG--  168
Q9D1Z9          100  DGICCPLREPCVLQKLGPLPGKDTP-----PDVTVPTSSLNKRLDSRLLFLDAPASGG--  152
Q60898          165  DGLCCWITTPCLAQNIPAPTSHPSPCTSPGSDVLRQKTFDVKRVSRLQEGSTG-TENPL  223
Q13239          165  DGLCCWLTTPCLTQSTAAPAVRAS-----SSDVLRQKTVDVRRVSRLQEDPSG-TENPL  218

NOV13           232  --EESLLSPGLRESLSPVPSLN-DEAVSLDDA---------------------------  261
Q9H6Q3          232  --EESLLSPGLRESLSPVPSLN-DEAVSLDDA---------------------------  261
Q9H135          168  --EESLLSPGLRESLSPVPSLN-DEAVSLDDA---------------------------  197
Q9D1Z9          152  --EASLLSPGLRESLSSVPSLA-EDP--LDDA---------------------------  179
Q60898          224  RVDESLPSYGLRESLTASVPSLTGDDSSSPLRKKKSLSLMYTGSKRKSSFFSAPQYFED  281
Q13239          219  GVDESLPSYGLRESLTASVPSLTSPDNTSPLRKKKSISLMYGGSKRKSSFFSSPPYFED  276
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Table 13F lists the domain description from DOMAIN analysis results against NOV13. This indicates that the NOV13 sequence has properties similar to those of other proteins known to contain this domain.

Table 13F. Domain Analysis of NOV13

PFAM Analysis

Model	Description	Score	E-value
SH2 (InterPro)	Src homology domain 2	110.5	4.6e-37
SH3 (InterPro)	SH3 domain	26.3	0.00012

PRODOM Analysis

Sequences producing High-scoring Segment Pairs:		High Score	Smallest Sum Probability
prdm:64	p36 (157) SRC(10) KSYK(8) YES(7) // DOMAIN KI...	214	2.4e-18
prdm:46	p36 (181) SRC(10) YES(7) GRB2(6) // DOMAIN SH...	77	0.0038

PROSITE Analysis

Pattern Name	Pattern	Number in NOV13
CAMP_PHOSPHO_SITE PS00004 (Interpro) PDOC00004	[RK]{2}.[ST]	2
PKC_PHOSPHO_SITE PS00005 (Interpro) PDOC00005	[ST].[RK]	6
CK2_PHOSPHO_SITE PS00006 (Interpro) PDOC00006	[ST].{2}[DE	4

BLOCKS Analysis

AC#	Description	Strength	Score
BL00512B	Alpha-galactosidase proteins.	1411	1054
BL00439A	Acyltransferases ChoActase/COT/ CPT	1390	1031
BL00543A	HlyD family secretion proteins.	1402	1029
BL00535B	Respiratory chain NADH dehydrogenase	1555	1025
BL00564G	Argininosuccinate synthase proteins.	1440	1023
BL01276C	Peptidase family U32 proteins.	1425	1023
BL00481F	Thiol-activated cytolysins proteins.	1675	1022
BL00117A	Galactose-1-phosph. uridyl transferase	1843	1020

Other BLAST results include sequences from the Patp database, which is a proprietary database that contains sequences published in patents and patent publications. Patp results include those listed in Table 13G.

Table 13G. Patp alignments of NOV13

Sequences producing High-scoring Segment Pairs:	High Score	Smallest Sum Prob. P(N)
patp:AAB42993 Human ORFX ORF2757 polypeptide sequence SEQ...	1269	3.0e-129
patp:AAY49420 PKA substrate, Src-family protein - Homo sa...	342	6.9e-31
patp:AAB37700 Human lymphocyte kinase - Homo sapiens, 508...	334	5.9e-30
patp:AAY29668 Human src-family kinase laloo protein - Hom...	300	3.8e-26
patp:AAY24421 Human yes1 protein - Homo sapiens, 543 aa.	300	3.8e-26

Receptor tyrosine kinases (RTKs) and their associated signaling pathways are critical to proper cell function, and perturbations in these pathways contribute to the onset and progression of diseases, *e.g.* cancer. Given the strong evidence that links signaling by certain families of RTKs to the progression of breast cancer, it is not surprising that the expression profile of key downstream signaling intermediates in this disease has also come under scrutiny, particularly because some exhibit transforming potential or amplify mitogenic signaling pathways when they are overexpressed. Reflecting the diverse cellular processes regulated by RTKs, it is now clear that altered expression of such signaling proteins in breast cancer may influence not only cellular

proliferation (*e.g.* Grb2) but also the invasive properties of the cancer cells (*e.g.* EMS1/cortactin).

SH2 domains are discrete structural motifs common to a variety of critical intracellular signaling proteins. Inhibitors of specific SH2 domains have become important therapeutic targets in the treatment and/or prevention of restenosis, cancers (including small cell lung), cardiovascular disease, osteoporosis, apoptosis among others. Considering the social and economic impact of these diseases significant attention has been focused on the development of potent and selective inhibitors of specific SH2 domains. In particular, considerable research has been performed on Src, PI 3-kinase, Grb2 and Lck.

Receptor tyrosine kinases are also important in diabetes. Diabetes mellitus is commonly considered as a disease of a scant beta-cell mass that fails to respond adequately to the functional demand. Tyrosine kinases may play a role for beta-cell replication, differentiation (neoformation) and survival. Transfection of beta-cells with DNA constructs coding for tyrosine kinase receptors yields a ligand-dependent increase of DNA synthesis in beta-cells. Several tyrosine kinase receptors, such as the VEGFR-2 (vascular endothelial growth factor receptor 2) and c-Kit, are present in pancreatic duct cells. Because ducts are thought to harbor beta-cell precursor cells, these receptors may play a role for the neoformation of beta-cells. The Src-like tyrosine kinase mouse Gtk (previously named Bsk/Iyk) is expressed in islet cells, inhibits cell proliferation. Furthermore, Gtk confers decreased viability in response to cytokine exposure. Shb is a Src homology 2 domain adaptor protein which participates in tyrosine kinase signaling. Transgenic mice overexpressing Shb in beta-cells exhibit an increase in the neonatal beta-cell mass, an improved glucose homeostasis, but also decreased survival in response to cytokines and streptozotocin. Thus, tyrosine kinase signaling may generate multiple responses in beta-cells, involving proliferation, survival and differentiation.

The disclosed NOV13 nucleic acid encoding a receptor tyrosine kinase-like protein includes the nucleic acid whose sequence is provided in Table 13A, or a fragment thereof. The invention also includes a mutant or variant nucleic acid any of whose bases may be changed from the corresponding base shown in Table 13A while still encoding a protein that maintains its receptor tyrosine kinase-like activities and physiological functions, or a fragment of such a nucleic acid. The invention further includes nucleic acids whose sequences are complementary to those just described, including nucleic acid fragments that are complementary to any of the nucleic acids just described. The invention additionally includes nucleic acids or nucleic acid fragments, or complements thereto, whose structures include chemical modifications. Such modifications include, by way of nonlimiting example, modified bases, and nucleic acids whose sugar phosphate backbones are modified or derivatized. These modifications are carried out at

least in part to enhance the chemical stability of the modified nucleic acid, such that they may be used, for example, as antisense binding nucleic acids in therapeutic applications in a subject

The disclosed NOV13 protein of the invention includes the receptor tyrosine kinase-like protein whose sequence is provided in Table 13B. The invention also includes a mutant or variant protein any of whose residues may be changed from the corresponding residue shown in Table 13B while still encoding a protein that maintains its receptor tyrosine kinase -like activities and physiological functions, or a functional fragment thereof.

The invention further encompasses antibodies and antibody fragments, such as F_{ab} or $(F_{ab})_2$ that bind immunospecifically to any of the proteins of the invention.

The above defined information for this invention suggests that this receptor tyrosine kinase -like protein (NOV13) may function as a member of a "receptor tyrosine kinase family". Therefore, the NOV13 nucleic acids and proteins identified here may be useful in potential therapeutic applications implicated in (but not limited to) various pathologies and disorders as indicated below. The potential therapeutic applications for this invention include, but are not limited to: cancer and diabetes research tools, for all tissues and cell types composing (but not limited to) those defined here, *e.g.* normal and cancerous tissue and pancreatic tissue.

Based on the tissues in which NOV13 is most highly expressed; including spleen and pituitary; specific uses include developing products for the diagnosis or treatment of a variety of diseases and disorders.

The NOV13 nucleic acids and proteins of the invention are useful in potential therapeutic applications implicated in cancer including but not limited to breast cancer and diabetes and/or other pathologies and disorders. For example, a cDNA encoding the receptor tyrosine kinase -like protein (NOV13) may be useful in cancer therapy, and the receptor tyrosine kinase-like protein (NOV13) may be useful when administered to a subject in need thereof. By way of nonlimiting example, the compositions of the present invention will have efficacy for treatment of patients suffering from cancer including but not limited to breast cancer. The NOV13 nucleic acid encoding receptor tyrosine kinase-like protein, and the receptor tyrosine kinase-like protein of the invention, or fragments thereof, may further be useful in diagnostic applications, wherein the presence or amount of the nucleic acid or the protein are to be assessed. Additional disease indications and tissue expression for NOV13 is presented in Example 2.

NOV13 nucleic acids and polypeptides are further useful in the generation of antibodies that bind immuno-specifically to the novel NOV13 substances for use in therapeutic or diagnostic methods. These antibodies may be generated according to methods known in the art, using prediction from hydrophobicity charts, as described in the "Anti-NOVX Antibodies" section below. The disclosed NOV13 protein has multiple hydrophilic regions, each of which

can be used as an immunogen. In one embodiment, a contemplated NOV13 epitope is from about amino acids 1 to 10. In another embodiment, a NOV13 epitope is from about amino acids 25 to 40. In additional embodiments, NOV13 epitopes are from about amino acids 100 to 110, from about amino acids 120 to 130 and from about amino acids 250 to 255. These novel proteins can be used in assay systems for functional analysis of various human disorders, which will help in understanding of pathology of the disease and development of new drug targets for various disorders.

NOV14

A disclosed NOV14 nucleic acid of 5193 nucleotides (also referred to as 87919652) encoding a novel multidrug resistance-associated transporter-like protein is shown in Table 14A. An open reading frame was identified beginning with an ATG initiation codon at nucleotides 71-73 and ending with a TGA codon at nucleotides 4652-4654. A putative untranslated region upstream from the initiation codon and downstream from the termination codon is underlined in Table 14A, and the start and stop codons are in bold letters.

Table 1A. NOV14 nucleotide sequence (SEQ ID NO:82).

CTCCGGCGCCCGCTCTGCCCGCCGCTGGGTCCGACCGCGCTCGCCTTCCTTGCAGCCGCGCCTCGGCCCCATGGACGCCC
TGTGCGGTTCCGGGGAGCTGGCTCCAAGTTCTGGGACTCCAACCTGTCTGTGCACACAGAAAACCCGGACCTCGCTCCC
TGCTTCCAGAACTCCCTGCTGGCCTGGGTGCCCTGCATCTACCTGTGGGTCGCCCTGCCCTGCTACTTGCTCTACCTGCG
GCACCATTGTCGTGGCTACATCATCCTCTCCCACCTGTCCAAGCTCAAGATGGTCCTGGGTGTCCTGCTGTGGTGCGTCT
CCTGGGCGGACCTTTTTTACTCCTTCCATGGCCTGGTCCATGGCCGGGCCCCTGCCCCTGTTTTCTTTGTCACCCCCTTG
GTGGTGGGGGTCACCATGCTGCTGGCCACCCTGCTGATACAGTATGAGCGGCTGCAGGGCGTACAGTCTTCGGGGGTCCT
CATTATCTTCTGGTTCCTGTGTGTGGTCTGCGCCATCGTCCCATTCCGCTCCAAGATCCTTTTAGCCAAGGCAGAGGGTG
AGATCTCAGACCCCTTCCGCTTCACCACCTTCTACATCCACTTTGCCCTGGTACTCTCTGCCCTCATCTTGGCCTGCTTC
AGGGAGAAACCTCCATTTTTCTCCGCAAAGAATGTCGACCCTAACCCCTACCCTGAGACCAGCGCTGGCTTTCTCTCCCG
CCTGTTTTTCTGGTGGTTCACAAAGATGGCCATCTATGGCTACCGGCATCCCCTGGAGGAGAAGACCTCTGGTCCCTAA
AGGAAGAGGACAGATCCCAGATGGTGGTGCAGCAGCTGCTGGAGGCATGGAGGAAGCAGGAAAAGCAGACGGCACGACAC
AAGGCTTCAGCAGCACCTGGGAAAAATGCCTCCGGCGAGGACGAGGTGCTGCTGGGTGCCGGCCCAGGCCCCGGAAGCC
CTCCTTCCTGAAGGCCCTGCTGGCCACCTTCGGCTCCAGCTTCCTCATCAGTGCCTGCTTCAAGCTTATCCAGGACCTGC
TCTCCTTCATCAATCCACAGCTGCTCAGCATCCTGATCAGGTTTATCTCCAACCCCATGGCCCCCTCCTGGTGGGGCTTC
CTGGTGGCTGGGCTGATGTTCCTGTGCTCCATGATGCAGTCGCTGATCTTACAACACTATTACCACTACATCTTTGTGAC
TGGGGTGAAGTTTCGTACTGGGATCATGGGTGTCATCTACAGGAAGGCTCTGGTTATCACCAACTCAGTCAAACGTGCGT
CCACTGTGGGGGAAATTGTCAACCTCATGTCAGTGGATGCCCAGCGCTTCATGGACCTTGCCCCCTTCCTCAATCTGCTG
TGGTCAGCACCCCTGCAGATCATCCTGGCGATCTACTTCCTCTGGCAGAACCTAGGTCCCTCTGTCCTGGCTGGAGTCGC
TTTCATGGTCTTGCTGATTCCACTCAACGGAGCTGTGGCCGTGAAGATGCGCGCGCTTCCAGGTAAAGCAAATGAAATTGA
AGGACTCGCGCATCAAGCTGATGAGTGAGATCCTGAACGGCATCAAGGTGCTGAAGCTGTACGCCTGGGAGCCCAGCTTC
CTGAAGCAGGTGGAGGGCATCAGGCAGGGTGAGCTCCAGCTGCTGCGCACGGCGGCCTACCTCCACACCACAACCACCTT
CACCTGGATGTGCAGCCCCTTCCTGGTGACCCTGATCACCCTCTGGGTGTACGTGTACGTGGACCCAAACAATGTGCTGG
ACGCCGAGAAGGCCTTTGTGTCTGTGTCCTTGTTTATATCTTAGACTTCCCCTCAACATGCTGCCCCAGTTAATCAGC
AACCTGACTCAGGCCAGTATGTCTCTGAAACGGATCCAGCAATTCCTGAGCCAAGAGGAACTTGACCCCAGAGTGTGGA
AAGAAAGACCATCTCCCCAGGCTATGCCATCACCATACACAGTGGCACCTTCACCTGGGCCCAGGACCTGCCCCCCACTC
TGCACAGCCTAGACATCCAGGTCCCGAAAGGGGCACTGGTGGCCGTGGTGGGGCCTGTGGGCTGTGGGAAGTCTCCCTG
GTGTCTGCCCTGCTGGAGAGATGGAGAAGCTAGAAGGCAAAGTGCACATGAAGGGCTCCGTGGCCTATGTGCCCCAGCA
GGCATGGATCCAGAACTGCACTCTTCAGGAAAACGTGCTTTTCGGCAAAGCCCTGAACCCCAAGCGCTACCAGCAGACTC
TGGAGGCCTGTGCCTTGCTAGCTGACCTGGAGATGCTGCCTGGTGGGGATCAGACAGAGATTGGAGAGAAGGGCATTAAC
CTGTCTGGGGCCAGCGGCAGCGGGTCAGTCTGGCTCGAGCTGTTTACAGTGATGCCGATATTTTCTTGCTGGATGACCC
ACTGTCCGCGGTGGACTCTCATGTGGCCAAGCACATCTTTGACCACGTCATCGGGCCAGAAGGCGTGCTGGCAGGCAAGA
CGCGAGTGCTGGTGACGCACGGCATTAGCTTCCTGCCCCAGACAGACTTCATCATTGTGCTAGCTGATGGGACAGGTGTCT
GAGATGGGCCCGTACCCAGCCCTGCTGCAGCGCAACGGCTCCTTTGCCAACTTTCTCTGCAACTATGCCCCCGATGAGGA
CCAAGGCACCTGGAGGACAGCTGGACCGCGTTGGAAGGTGCAGAGGATAAGGAGCACTGCTGATTGAAGACACACCTCA
GCAACCACACGGATCTGACAGACAATGATCCAGTCACCTATGTGGTCCAGAAGCAGTTTATGAGACAGCTGAGTGCCCTG
TCCTCAGATGGGGAGGGACAGGGTCGGCCTGTACCCCGGAGCACCTGGGTCCATCAGAGAAGGTGCAGGTGACAGAGGC
GAAGGCAGATGGGGCACTGACCCAGGAGGAGAAAGCAGCCATTGGCACTGTGGAGCTCAGTGTGTTCTGGGATTATGCCA

AGGCCGTGGGGCTCTGTACCACGCTGGCCATCTGTCTCCTGTATGTGGGTCAAAGTGCGGCTGCCATTGGAGCCAATGTG
TGGCTCAGTGCCTGGACAAATGATGCCATGGCAGACAGTAGACAGAACAACACTTCCCTGAGGCTGGGCGTCTATGCTGC
TTTAGGAATTCTGCAAGGGTTCTTGGTGATGCTGGCAGCCATGGCCATGGCAGCGGGTGGCATCCAGGCTGCCCGTGTGT
TGCACCAGGCACTGCTGCACAACAAGATACGCTCGCCACAGTCCTTCTTTGACACCACACCATCAGGCCGCATCCTGAAC
TGCTTCTCCAAGGACATCTATGTCGTTGATGAGGTTCTGGCCCCTGTCATCCTCATGCTGCTCAATTCCTTCTTCAACGC
CATCTCCACTCTTGTGGTCATCATGGCCAGCACGCCGCTCTTCACTGTGGTCATCCTGCCCCTGGCTGTGCTCTACACCT
TAGTGCAGCGCTTCTATGCAGCCACATCACGGCAACTGAAGCGGCTGGAATCAGTCAGCCGCTCACCTATCTACTCCCAC
TTTTCGGAGACAGTGACTGGTGCCAGTGTCATCCGGGCCTACAACCGCAGCCGGGATTTTGAGATCATCAGTGATACTAA
GGTGGATGCCAATCAGAGAAGCTGCTACCCCTACATCATCTCCAACCGGTGGCTGAGCATCGGAGTGGAGTTCGTGGGGA
ACTGCGTGGTGCTCTTTGCTGCACTATTTGCCGTCATCGGGAGGAGCAGCCTGAACCCGGGGCTGGTGGGCCTTTCTGTG
TCCTACTCCTTGCAGGTGACATTTGCTCTGAACTGGATGATACGAATGATGCCAGATTTGGAATCTAACATCGTGGCTGT
GGAGAGGGTCAAGGAGTACTCCAAGACAGAGACAGAGGCGCCCTGGGTGGTGGAAGGCAGCCGCCCTCCCGAAGGTTGGC
CCCCACGTGGGGAGGTGGAGTTCCGGAATTATTCTGTGCGCTACCGGCCGGGCCTAGACCTGGTGCTGAGAGACCTGAGT
CTGCATGTGCACGGTGGCGAGAAGGTGGGGATCGTGGGCCGCACTGGGGCTGGCAAGTCTTCCATGACCCTTTGCCTGTT
CCGCATCCTGGAGGCGGCAAAGGGTGAAATCCGCATTGATGGCCTCAATGTGGCAGACATCGGCCTCCATGACCTGCGCT
CTCAGCTGACCATCATCCCGCAGGACCCCATCCTGTTCTCGGGGACCCTGCGCATGAACCTGGACCCCTTCGGCAGCTAC
TCAGAGGAGGACATTTGGTGGGCTTTGGAGCTGTCCCACCTGCACACGTTTGTGAGCTCCCAGCCGGCAGGCCTGGACTT
CCAGTGCTCAGAGGGCGGGGAGAATCTCAGCGTGGGCCAGAGGCAGCTGGTGTGCCTGGCCCGAGCCCTGCTCCGCAAGA
GCCGCATCCTGGTTTTAGACGAGGCCACAGCTGCCATCGACCTGGAGACTGACAACCTCATCCAGGCTACCATCCGCACC
CAGTTTGATACCTGCACTGTCCTGACCATCGCACACCGGCTTAACACTATCATGGACTACACCAGGGTCCTGGTCCTGGA
CAAAGGAGTAGTAGCTGAATTTGATTCTCCAGCCAACCTCATTGCAGCTAGAGGCATCTTCTACGGGATGGCCAGAGATG
CTGGACTTGCCTAAAATATATTCCTGAGATTTCCTCCTGGCCTTTCCTGGTTTTCATCAGGAAGGAAATGACACCAAATA
TGTCCGCAGAATGGACTTGATAGCAAACACTGGGGGCACCTTAAGATTTTGCACCTGTAAAGTGCCTTACAGGGTAACTG
TGCTGAATGCTTTAGATGAGGAAATGATCCCCAAGTGGTGAATGACACGCCTAAGGTCACAGCTAGTTTGAGCCAGTTAG
ACTAGTCCCCGGTCTCCCGATTCCCAACTGAGTGTTATTTGCACACTGCACTGTTTTCAAATAACGATTTTATATGAAATGA
CCTCTGTCCTCCCTCTGATTTTTCATATTTTCTAAAGTTTCGTTTCTGTTTTTTAATAAAAAGCTTTTTCCTCCTGGAAC
AGAAGACAGCTGCTGGGTCAGGCCACCCCTAGGAACTCAGTCCTGTACTCTGGGGTGCTGCCTGAATCCATTAAAAATGG
GAGTACTGATGAAATAAAACTACATGGTCAACAGTAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAA

The NOV14 nucleic acid was identified on chromosome 17 by comparing it to the human genome sequence. Exons were predicted by homology and the intron/exon boundaries were determined using standard genetic rules. Exons were further selected and refined by means of similarity determination using multiple BLAST (for example, tBlastN, BlastX, and BlastN) searches, and, in some instances, GeneScan and Grail. Expressed sequences from both public and proprietary databases were also added when available to further define and complete the gene sequence. The DNA sequence was then manually corrected for apparent inconsistencies thereby obtaining the sequences encoding the full-length protein. The NOV14 nucleic acid was further mapped to the 17q21 locus. This locus is associated with breast cancer (OMIM 176705, 113705), glycogen storage disease (OMIM 232200), essential hypertension (OMIM 171190) and/or other diseases/disorders.

In a search of public sequence databases, the NOV14 nucleic acid sequence has 5151 of 5155 bases (99%) identical to a human ATP-binding cassette, sub-family C (Accession No. XM_038002). Public nucleotide databases include all GenBank databases and the GeneSeq patent database.

A disclosed NOV14 polypeptide (SEQ ID NO:83) encoded by SEQ ID NO:82 has 1527 amino acid residues and is presented in Table 14B using the one-letter amino acid code. SignalP, Psort and/or Hydropathy results predict that NOV14 has a signal peptide and is likely to be localized to the plasma membrane with a certainty of 0.8000. The most likely cleavage site for a NOV14 peptide is between amino acids 53 and 54 of SEQ ID NO.28, *i.e.* at CYL-LY.

Table 14B. Encoded NOV14 protein sequence (SEQ ID NO:83).

MDALCGSGELGSKFWDSNLSVHTENPDLTPCFQNSLLAWVPCIYLWVALPCYLLYLRHHCRGYIILSHLSKLKMVLGVLLW
CVSWADLFYSFHGLVHGRAPAPVFFVTPLVVGVTMLLATLLIQYERLQGVQSSGVLIIFWFLCVVCATVPFRSKILLAKAE
GEISDPFRFTTFYIHFALVLSALILAACFREKPPFFSAKNVDPNPYPETSAGFLSRLFFWWFTKMAIYGYRHPLEEKDLWSL
KEEDRSQMVVQQLLEAWRKQEKQTARHKASAAPGKNASGEDEVLLGARPRPRKPSFLKALLATFGSSFLISACFKLIQDLL
SFINPQLLSILIRFISNPMAPSWWGFLVAGLMFLCSMMQSLILQHYVHYIFVTGVKFRTGIMGVIYRKALVITNSVKRAST
VGEIVNLMSVDAQRFMDLAPFLNLLWSAPLQIILAIYFLWQNLGPSVLAGVAFMVLLIPLNGAVAVKMRAFQVKQMKLKDS
RIKLMSEILNGIKVLKLYAWEPSFLKQVEGIRQGELQLLRTAAYLHTTTFTWMCSPFLVTLITLWVYVVDPNNVLDAEK
AFVSVSLFNILRLPLNMLPQLISNLTQASVSLKRIQQFLSQEELDPQSVERKTISPGYAITIHSGTFTWAQDLPPTLHSLD
IQVPKGALVAVVGPVGCGKSSLVSALLGEMEKLEGKVHMKGSVAYVPQQAWIQNCTLQENVLFGKALNPKRYQQTLEACAL
LADLEMLPGGDQTEIGEKGINLSGGQRQRVSLARAVYSDADIFLLDDPLSAVDSHVAKHIFDHVIGPEGVLAGKTRVLVTH
GISFLPQTDFIIVLADGQVSEMGPYPALLQRNGSFANFLCNYAPDEDQGHLEDSWTALEGAEDKEALLIEDTLSNHTDLTD
NDPVTYVVQKQFMRQLSALSSDGEQGGRPVPRRHLGPSEKVQVTEAKADGALTQEEKAAIGTVELSVFWDYAKAVGLCTTL
AICLLYVGQSAAAIGANVWLSAWTNDAMADSRQNNTSLRLGVYAALGILQGFLVMLAAAMAAAGGIQAARVLHQALLHNKI
RSPQSFFDTTPSGRILNCFSKDIYVVDEVLAPVILMLLNSFFNAISTLVVIMASTPLFTVVILPLAVLYTLVQRFYAATSR
QLKRLESVSRSPIYSHFSETVTGASVIRAYNRSRDFEIISDTKVDANQRSCYPYIISNRWLSIGVEPVGNCVVLFAALFAV
IGRSSLNPGLVGLSVSYSLQVTFALNWMIRMMSDLESNIVAVERVKEYSKTETEAPWVVEGSRPPEGWPPRGEVEFRNYSV
RYRPGLDLVLRDLSLHVHGGEKVGIVGRTGAGKSSMTLCLFRILEAAKGEIRIDGLNVADIGLHDLRSQLTIIPQDPILFS
GTLRMNLDPFGSYSEEDIWWALELSHLHTFVSSQPAGLDFQCSEGGENLSVGQRQLVCLARALLRKSRILVLDEATAAIDL
ETDNLIQATIRTQFDTCTVLTIAHRLNTIMDYTRVLVLDKGVVAEFDSPANLIAARGIFYGMARDAGLA

In a search of public sequence databases, the NOV14 amino acid sequence has 1527 of 1527 amino acid residues (100%) identical to, and 1527 residues (100%) positive with, the 1527 amino acid residue human canicular multispecific organic anion transporter/multidrug resistance-associated protein (Accession No. O15438). Public amino acid databases include the GenBank databases, SwissProt, PDB and PIR. It was also found that NOV14 had homology to the amino acid sequences shown in the BLASTP data listed in Table 14C.

Table 14C. BLAST results for NOV14

Gene Index/ Identifier	Protein/ Organism	Length (aa)	Identity (%)	Positives (%)	Expect
MRP3_HUMAN; O15438; BAA28146.1; CAA76658.1; CAA76658.1; AAD01430.1;	CANALICULAR MULTISPECIFIC ORGANIC ANION TRANSPORTER 2 (MULTIDRUGRESISTANCE-ASSOCIATED PROTEIN 3). homo sapiens. 5/2000	1527	1527/1527 (100%)	1527/1527 (100%)	0.0
MRP3_RAT; O88563; AAC25416.1; BAA28955.1	CANALICULAR MULTISPECIFIC ORGANIC ANION TRANSPORTER 2 (MULTIDRUGRESISTANCE-ASSOCIATED PROTEIN 3) (MRP-LIKE PROTEIN-2) (MLP-2). rattus norvegicus. 5/2000	1522	1194/1528 (78%)	1334/1528 (87%)	0.0
MRP1_HUMAN; P33527; AAB46616.1; AAB83983.1	MULTIDRUG RESISTANCE-ASSOCIATED PROTEIN 1. homo sapiens. 5/2000	1531	872/1538 (57%)	1131/1538 (74%)	0.0
Q9UQ99; AF022853; AAB83979.1	MULTIDRUG RESISTANCE PROTEIN (FRAGMENT). homo sapiens. 6/2001	1515	870/1529 (57%)	1128/1529 (74%)	0.0
O35379; AF022908; AAB80938.1	MULTIDRUG RESISTANCE PROTEIN. mus musculus. 6/2001	1528	859/1540 (56%)	1117/1540 (73%)	0.0

The alignment and homology of these sequences is shown graphically in the ClustalW analysis in Table 14D.

Table 14D Information for the ClustalW proteins

1) NOV14 (SEQ ID NO:83)
2) MRP3_HUMAN (SEQ ID NO:84)
3) MRP3_RAT (SEQ ID NO:85)
4) MRP1_HUMAN (SEQ ID NO:86)
5) Q9UQ99 (SEQ ID NO:87)
6) O35379 (SEQ ID NO:88)

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NOV14            1   -MDALCGSGELGSKFWDSNLSVHTDNPLTTCFQNSLLLWVPCTYLWVALCYLLVLRHL  59
MRP3_HUMAN       1   -MDALCGSGELGSKFWDSNLSVHTDNPLTTCFQNSLLLWVPCTYLWVALCYLLVLRHL  59
MRP3_RAT         1   -MDRLCGSGELGSKFWDSNLTVYTDNPLTTCFQNSLLLWVPCTYLWVALCYLLVLRHL  59
MRP1_HUMAN       1   MALRGFCLVGSDPLWDWNVTWNTDNPLTTCFQNTLLWVPCTYLWVACFTYFLVLSRL  60
Q9UQ99           1   ----------------DWNVTWNTDNPLTTCFQNTLLWVPCTYLWVACFTYFLVLSRL  44
O35379           1   MALRSFCSADGSDPLWDWNVTWHTDNPLTTCFQNTLLWVPCTYLWVSCFTYFLVLSRL  60

NOV14           60   CRGVTLTSHLSLLKWVGVLLWCVSWADLFYSFHGLVHCRAPAPVPVFLVVGVLVLLA  119
MRP3_HUMAN      60   CRGVTLTSHLSLLKWVGVLLWCVSWADLFYSFHGLVHCRAPAPVPVFLVVGVLVLLA  119
MRP3_RAT        60   RLGVTLTSHLSLLKWVGVLLWCISWADLFYSFHGLVHCSSPAPVPVFLVVGVLVLLA  119
MRP1_HUMAN      61   LRGVTLWTHLSLLKWTNVLGLLWVCWADLFYSFWERSRCIFLAPVPVFLVSGTLLVLLA  120
Q9UQ99          45   DRGVTLWTHLSLLKWTNVLGLLWVCWADLFYSFWERSRCIFLAPVPVFLVSGTLLVLLA  104
O35379          61   DRGVTLWTHLSLLKWTNALGFLWMICWADLFYSFWERSQCVLRAPVLLVSPTLLSLVLLA  120

NOV14           120  TLLTQVEELQGVQSSGVLTTFWFLCVVCAIVPRSKLLAKAEGEISLPPRPTTFWIPFA  179
MRP3_HUMAN      120  TLLTQVEELQGVQSSGVLIIFWFLCVVCAIVPRSKLLAKAEGEISLPPRPTTFWIPFA  179
MRP3_RAT        120  TLLTQVEELQGVESSGVLIIFWFLCWICAIIPRSKLLAAAEGKILLPPRPTTFWIPFA  179
MRP1_HUMAN      121  TLLTQVEERKGVQSSGIMLTFWFLVALVCALAILKSKIWTALKEDAQVVLPRTLFVWYFS  180
Q9UQ99          105  TLLTQVEERKGVQSSGIMLTFWFLVALVCALAILKSKIWTALKEDAQVVLPRTLFVWYFS  164
O35379          121  TLLTQVEERKGVQSSGIMLTFWFLVALLCALAILKSKIESALKKDAHVVVPRTSFFWLFT  180

NOV14           180  TVLSAILIAGCREKPWTPSAKNVLTNFVPFSATGFLSRGFFWWFTKMAIYGYRSPLESKT  239
MRP3_HUMAN      180  TVLSAILIAGCREKPWTPSAKNVLTNFVPFSATGFLSRGFFWWFTKMAIYGYRSPLESKT  239
MRP3_RAT        180  TVLSAILIAGCREKPWTPSAKNVLTNFVPFSATGFLSRGFFWWFTKMALYGYRSPLESKT  239
MRP1_HUMAN      181  LLLIQVESSCFSDRSCLFSETIHFNNPCPFSASGFLSRGFFWWFTGLIVRGYRQPLESSI  240
Q9UQ99          165  LLLIQVESSCFSDRSCLFSETIHFNNPCPFSASGFLSRGFFWWFTGLIVRGYRQPLESSI  224
O35379          181  LMLVQVESSCFSDCSCLFSETVHFNNPCPFSASGFLSRGFFWWFTGMMVRGYRQPLESSI  240

NOV14           240  LWSLKEDPSQGMVVQCLEAWKQEKQTARHKASAAPGK-------------NASGPEGV  286
MRP3_HUMAN      240  LWSLKEDPSQGMVVQCLEAWKQEKQTARHKASAAPGK-------------NASGPEGV  286
MRP3_RAT        240  LWSLSEDLCCHRVVQRLEAWKQQQTQAS-GPQTAALEP-------------KIAGPDGV  285
MRP1_HUMAN      241  LWSLNKEDTSEQVVPVFVKNWKKECAKTRKQPVKVVYSS-KDPAQPKESSKVDANEPVDA  299
Q9UQ99          225  LWSLNKEDTSEQVVPVFVKNWKKECAKTRKQPVKVVYSS-KDPAQPKESSKVDANEPVDA  283
O35379          241  LWSLNKELTSEPVVPVFVNNWKKECDKSRKQPVRIVYAPPKDPSKPKGSSQLDVNEPVDA  300

NOV14           287  LEGARS-RPRKPSFLKQLTAMFGSSFLISAGFKLIQDLLSFVNPQLLSILIRFTSNPMTP  345
MRP3_HUMAN      287  LEGARS-RPRKPSFLKQLTAMFGSSFLISAGFKLIQDLLSFVNPQLLSILIRFTSNPMTP  345
MRP3_RAT        286  LEKARS-RTKKPSFLRALVRTFTSSLIMGAGFKLIQDLSPSS-THSCSASSSGDFRPHGP  343
MRP1_HUMAN      300  LIVKSPQKEWNPSLFKVLYKTFGPYFLMSFFEAAHFLPDLMSGSGPQIKLLIKFVNDTKP  359
Q9UQ99          284  LIVKSPQKEWNPSLFKVLYKTFGPYFLMSFFEAAHFLPDLMSGSGPQIKLLIKFVNDTKP  343
O35379          301  LIVKSPHDRDPSLFKVLYKTFGPYFLNSFLYAALHLDMMSGAGPKILLDLIHNFVDRERP  360

NOV14           346  SMWSFLVAGLLCSMMQSLIFQHVMYHIFVWCVKFFTGIMCVIYRKALITTNSVKRASTL  405
MRP3_HUMAN      346  SMWSFLVAGLLCSMMQSLIFQHVMYHIFVWCVKFFTGIMCVIYRKALITTNSVKRASTL  405
MRP3_RAT        344  YWWSFLLAGLWVSSTMQLLIFHQHYHCIFVMALRIFTAICVIYRKALITTNSVKREVTL  403
MRP1_HUMAN      360  DWQSYFYTVPLLVTACLLGLVFHQVVHQHICPVSQMRIFTAVWCAVYRKALVITNSARKSST  419
Q9UQ99          344  DWQSYFYTVPLLVTACLLGLVFHQVVHQHICPVSQMRIFTAVWCAVYRKALVITNSARKSST  403
O35379          361  DWQSYFYTAPLLVSACLLGLVAHQVQHQHICPVSQMRIFTAVWCAVYRKALVITNTARKSST  420

NOV14           406  VGEIVNLMSVDAQRFMDLLPFLNLLWSAPLQIILALVFLWQNLGPSVLAGVAVMVLLTPL  465
MRP3_HUMAN      406  VGEIVNLMSVDAQRFMDLLPFLNLLWSAPLQIILALVFLWQNLGPSVLAGVAVMVLLTPL  465
MRP3_RAT        404  VGEVVNLMSVDAQRFMDVSPFLNLLWSAPLQIILALVFLWQLGPSVLAGVAVMVLLTPL  463
MRP1_HUMAN      420  VGEIVNLMSVDAQRFMDLLTYLNMIWSAPLQVILALYLLWLNLGPSVLAGVAVMVVLVPV  479
Q9UQ99          404  VGEIVNLMSVDAQRFMDLLTYLNMIWSAPLQIILALYLLWLNLGPSVLAGVAVMVVLVPV  463
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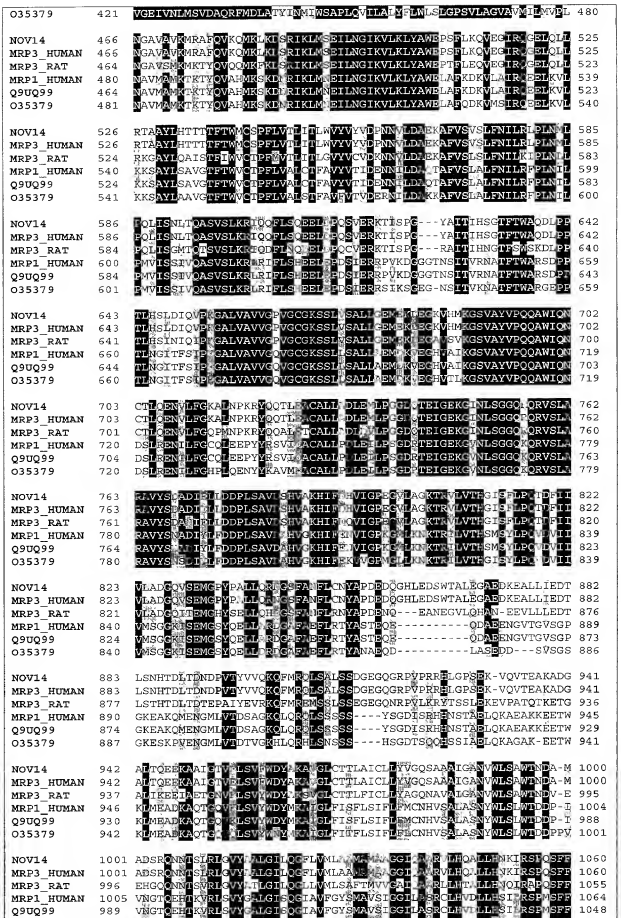


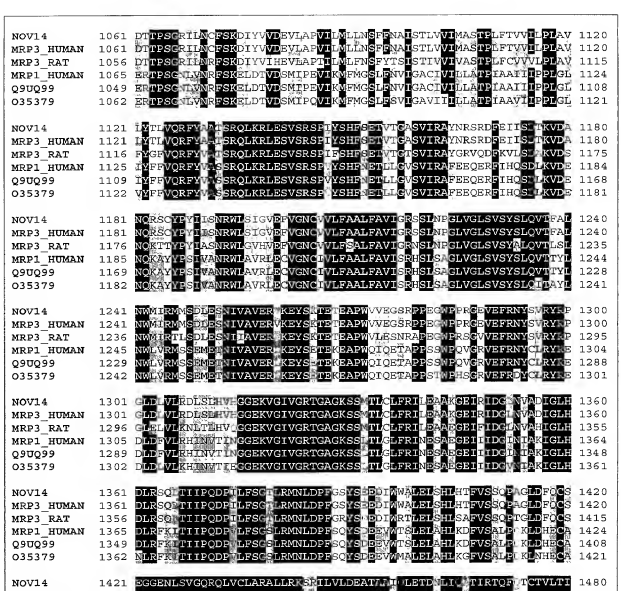


Table 14E lists the domain description from DOMAIN analysis results against NOV14. This indicates that the NOV14 sequence has properties similar to those of other proteins known to contain this domain.

Table 14E. Domain Analysis of NOV14

```
PROSITE
Pattern Name
LEUCINE_ZIPPER PS00029 (Interpro) PDOC00029    2 positions in NOV14
ABC_TRANSPORTER PS00211 (Interpro) PDOC00185   2 sites in NOV14

PRODOM                                                          Smallest Sum
                                                        High    Probability
Sequences producing High-scoring Segment Pairs:         Score     P(N)
prdm:8775  p36 (3) MRP2(2) MRP1(1)  - MULTIDRUG PROTEIN ...  384    7.1e-35
prdm:1070  p36 (21) CFTR(7) SUR(3) MRP2(2)  - TRANSMEMBR...  305    1.9e-26
prdm:923   p36 (24) CFTR(7) MRP2(4) SUR(3)  - TRANSMEMBR...  244    5.8e-20
prdm:993   p36 (22) CFTR(7) SUR(3) MRP2(2)  - TRANSMEMBR...  214    9.0e-17

BLOCKS
AC#       Description                                  Strength  Score
BL00211B  ABC transporters family proteins.             1331    1326
BL01247C  Inosine-uridine preferring nucleoside hydrola 1351    1084
BL00577B  Avidin / Streptavidin family proteins.        1442    1067
BL00853E  Beta-eliminating lyases pyridoxal-phosphate a 1602    1064
BL00019E  Actinin-type actin-binding domain proteins.   1179    1060
BL00256   Adipokinetic hormone family proteins.         1358    1057
BL00545B  Aldose 1-epimerase  proteins.                 1282    1056
BL00699A  Nitrogenases component 1 alpha and beta subun 1357    1056
```

Other BLAST results include sequences from the Patp database, which is a proprietary database that contains sequences published in patents and patent publications. Patp results include those listed in Table 1F.

Table 1F. Patp alignments of NOV14

Sequences producing High-scoring Segment Pairs:	High Score	Smallest Sum Prob. P(N)
patp:AAY43543 A human MPR-related ABC transporter designa...	7845	0.0
patp:AAW33363 Human multidrug resistance-associated prote...	7679	0.0
patp:AAR54928 Multidrug resistance protein - Homo sapiens...	4470	0.0
patp:AAR93153 Multi-drug resistance protein - Homo sapien...	4470	0.0
patp:AAW57485 Human multidrug resistance-associated prote...	4470	0.0

Members of the multidrug resistance-associated transporter-like protein family are critical modulators of cell physiology, and perturbations are associated with many diseases/disorders. Multidrug resistance (MDR) describes the phenomenon of simultaneous resistance to unrelated drugs. The two MDR genes identified in humans to date (the MDR-associated protein (MRP) and Pgp genes) are structurally similar and both are members of the ATP-binding cassette (ABC) transporter family. Although the physiological role of MRP is not yet understood, one Pgp gene (mdr1) plays an important role in the blood-tissue barrier and the other (mdr2/3) is involved in phospholipid transport in the liver. A variety of compounds (chemosensitizing agents) can interfere with Pgp and MRP function; such agents may improve the efficacy of conventional therapy when used in combination with such regimens. Determining the roles cellular MDR mechanisms play in patients' response to chemotherapy is a major

challenge. Using Pgp and MRP as molecular markers to detect MDR tumor cells is technically demanding, and solid tumors in particular contain heterogeneous cell populations. Since MDR requires Pgp or MRP gene expression, clinically relevant gene expression thresholds need to be established; sequential samples from individual patients are valuable for correlating MDR gene expression with the clinical course of disease. Studies in leukemias, myelomas, and some childhood cancers show that Pgp expression correlates with poor response to chemotherapy. However, in some cases, inclusion of a reversing or chemosensitizing agent such as verapamil or cyclosporin A has improved clinical efficacy. Such agents may inactivate Pgp in tumor cells or affect Pgp function in normal cells, resulting in altered pharmacokinetics. The ABC transporter superfamily in prokaryotes and eukaryotes is involved in the transport of substrates ranging from ions to large proteins. Of the 15 or more ABC transporter genes characterized in human cells, two (Pgp and MRP) cause MDR. Therefore, it would be relevant to determine the number of such genes present in the human genome; however, extrapolating from the number of ABC transporter genes in bacteria, the human gene probably contains a minimum of 200 ABC transporter superfamily members. Thus, tumor cells can potentially use many ABC transporters to mount resistance to known and future therapeutic agents.

Members of the multidrug resistance-associated transporter-like protein family are also important in liver disease. In several liver diseases the biliary transport is disturbed, resulting in, for example, jaundice and cholestasis. Many of these symptoms can be attributed to altered regulation of hepatic transporters. Organic anion transport, mediated by the canalicular multispecific organic anion transporter (cmoat), has been extensively studied. The regulation of intracellular vesicular sorting of CMOAT by protein kinase C and protein kinase A, and the regulation of cmoat-mediated transport in endotoxemic liver disease, have been examined. The discovery that the multidrug resistance protein (MRP), responsible for multidrug resistance in cancers, transports similar substrates as cmoat led to the cloning of a MRP homologue from rat liver, named mrp2. Mrp2 turned out to be identical to cmoat. At present there is evidence that at least two mrp's are present in hepatocytes, the original mrp (mrp1) on the lateral membrane, and mrp2 (cmoat) on the canalicular membrane. The expression of mrp1 and mrp2 in hepatocytes appears to be cell-cycle-dependent and regulated in a reciprocal fashion. These findings show that biliary transport of organic anions and possibly other canalicular transport is influenced by the entry of hepatocytes into the cell cycle.

Further, members of the multidrug resistance-associated transporter-like protein family are involved in various leukaemias. Approximately 15-30% of acute myeloid leukaemia (AML) patients are primarily resistant to chemotherapy, and 60-80% of patients who achieve complete remission will inevitably relapse and succumb to their disease. The multidrug resistant (MDR)

phenotype has been suspected as a major mechanism of therapy failure in AML; it is one of the best understood mechanisms of resistance to anticancer drugs. The classical MDR phenotype is characterized by the reduced ability of cells to accumulate drugs as compared to normal cells. The increased drug efflux is due to the activity of a 170 kDa glycoprotein, the P-glycoprotein (Pgp), a unidirectional drug-efflux pump which is encoded by the MDR1 gene. While studies of myeloid leukaemia and myeloma have provided the best evidence for the potential association between Pgp expression and clinical outcome, the lack of standardized methods for MDR detection and perhaps even more importantly, inconsistencies in the interpretation of MDR expression data account for divergent results in the literature. The clinicians' strong interest in MDR stems from the availability of agents capable of interfering with MDR, at least in vitro. If these laboratory results were reproducible in vivo, reversal of MDR would offer a rare opportunity to incorporate laboratory experience into the clinical management of patients.

The NOV14 nucleic acids are useful for screening a test compound for inhibition of MDR mediated transport, indicated by restoration of anticancer drug sensitivity, which in turn causes a reduction of transporter mediated cellular efflux of anticancer agents. The disclosed NOV14 nucleic acid encoding a multidrug resistance-associated transporter-like protein includes the nucleic acid whose sequence is provided in Table 14A, or a fragment thereof. The invention also includes a mutant or variant nucleic acid any of whose bases may be changed from the corresponding base shown in Table 14A while still encoding a protein that maintains its multidrug resistance-associated transporter -like activities and physiological functions, or a fragment of such a nucleic acid.

The disclosed NOV14 protein of the invention includes the multidrug resistance-associated transporter -like protein whose sequence is provided in Table 14B. The invention also includes a mutant or variant protein any of whose residues may be changed from the corresponding residue shown in Table 14B while still encoding a protein that maintains its multidrug resistance-associated transporter -like activities and physiological functions, or a functional fragment thereof.

The above defined information for this invention suggests that this multidrug resistance-associated transporter -like protein (NOV14) may function as a member of a "multidrug resistance-associated transporter family". Therefore, the NOV14 nucleic acids and proteins identified here may be useful in potential therapeutic applications implicated in (but not limited to) various pathologies and disorders as indicated below. The potential therapeutic applications for this invention include, but are not limited to: cancer and liver disease research tools, for all tissues and cell types composing (but not limited to) those defined here, *e.g.* cancerous and

normal tissue and liver tissue. Additional disease indications and tissue expression for NOV14 is presented in Example 2.

The NOV14 nucleic acids and proteins of the invention are useful in potential therapeutic applications implicated in cancer including but not limited to cancer, liver disease and/or other pathologies and disorders. For example, a cDNA encoding the multidrug resistance-associated transporter -like protein (NOV14) may be useful in liver disease therapy, and the multidrug resistance-associated transporter-like protein (NOV14) may be useful when administered to a subject in need thereof. By way of nonlimiting example, the compositions of the present invention will have efficacy for treatment of patients suffering from liver disease and cancer including but not limited to leukemia. The NOV14 nucleic acid encoding multidrug resistance-associated transporter -like protein, and the multidrug resistance-associated transporter -like protein of the invention, or fragments thereof, may further be useful in diagnostic applications, wherein the presence or amount of the nucleic acid or the protein are to be assessed.

NOV14 nucleic acids and polypeptides are further useful in the generation of antibodies that bind immuno-specifically to the novel NOV14 substances for use in therapeutic or diagnostic methods. These antibodies may be generated according to methods known in the art, using prediction from hydrophobicity charts, as described in the "Anti-NOVX Antibodies" section below. The disclosed NOV14 protein has multiple hydrophilic regions, each of which can be used as an immunogen. In one embodiment, a contemplated NOV14 epitope is from about amino acids 200 to 300. In another embodiment, a NOV14 epitope is from about amino acids 300 to 400. In additional embodiments, NOV14 epitopes are from about amino acids 900 to 300 and from about amino acids1400 to 1500. These novel proteins can be used in assay systems for functional analysis of various human disorders, which will help in understanding of pathology of the disease and development of new drug targets for various disorders.

NOV15

NOV15 includes two novel novel intracellular thrombospondin domain containing protein-like proteins disclosed below. The disclosed proteins have been named NOV15a and NOV15b.

NOV15a

A disclosed NOV15a nucleic acid of 1794 nucleotides (also referred to as 100399281 and 159518754) encoding a novel thrombospondin-like protein is shown in Table 15A. A partial open reading frame was identified beginning with an GGA codon at nucleotides 178-180 and ending with a TAA codon at nucleotides 1792-1794. A putative untranslated intronic region

upstream from the first in-frame coding triplet is underlined in Table 15A, and the start and stop codons are in bold letters.

Table 15A. NOV15a Nucleotide Sequence (SEQ ID NO:89)

ACGCGTAGCCACAAGACCGGGTCCGTTTCTGGTTGCCGTTCCCGCAGGTGACGCTGCAGACAGACCAGAGACTCCAGTC
ACCCTCGCCATCTGTGGAATCATATTCTGGCTGATCTTTGGTTTCAAAAGTCCGGTGGGCTGGGGCTGTATGGTCCCAC
CCCCTGGGGGGGTTGAGGAAGTTGCTGTCGTCTGAGGTACTGCCGTACGTGTAGTCCTGAAACCAGCTTTTCTCTCTCC
AAAGAAGCACCAAGGGAGCATCTGGACCACCAAGCTGCACACCAACCCTTCCCCAGACCGCGATTCCGACAAGAGACGG
GGCACCCTTCATTGCAAAGAGATTTCCCCAGATCCTTTCTCCTTGATCTACCAAACTTTCCAGATCTTTCCAAAGCTGA
TATCAATGGGCAGAATCCAAATATCCAGGTCACCATAGAGGTGGTCGACGGTCCTGACTCTGAAGCAGATAAAGATCAG
CATCCGGAGAATAAGCCCAGCTGGTCAGTCCCATCCCCCGACTGGCGGGCCTGGTGGCAGAGGTCCCTGTCCTTGGCCA
GGGCAAACAGCGGGGACCAGGACTACAAGTACGACAGTACCTCAGACGACAGCAACTTCCTCAACCCCCCCAGGGGGTG
GGACCATACAGCCCCAGGCCACCGGACTTTGAAACCAAAGATCAGCCAGAATATGATTCCACAGATGGCGAGGGTGAC
TGGAGTCTCTGGTCTGTCTGCAGCGTCACCTGCGGGAACGGCAACCAGAAACGGACCCGGTCTTGTGGCTACGCGTGCA
CTGCAACAGAATCGAGGACCTGTGACCGTCCAAACTGCCCAGCTTGCACCGGATTCCTGATTGTAAAGGAAGCTTGGTT
AGGGGTGGTAGTTTGGCATGTCCCTGCACCTCCAACTGGCAACCCCTCTGTGCCTTTGCCTGAGGTCTTTCTCTGGACC
CGAGCCCAGCTGCGCATGAATGCACAGGGCATTCCTAGCTGGAAATCCAGGACCAGTCCCCTGTCAGTGATGAATGGGA
GCTGGTGGATAAAAACTCAGATCCCCATCAATAAAAACAAATCCGGACTCAGTAAGGAGAGGATTTATTCAAAGGATTA
TTGCAGGGAGGCAAGGGATGTTATCTCCCTATTATTGCAATGGGATGAACGCTGTGACCATAAGATCTGCAAGCATCTC
AAGGAACAGCCTGGTGTCACATGTCCTTGAAGCACCTCCTGTGGGCCGGTTGTACACGCGGTGAGAGGGTTCTCTTT
GGCCTTTTCCAGACACAGACAGCTGTGAGCGCTGGATGAGCTTCAAAGCGAGGTCTTAAAGAAGTACATGCACAAGGT
GATGAATGACCTGCCCAGCTGCCCCTGCTCCTACCCCACTGAGGTGGCCTACAGCACGGCGGACATCTTCGACCGCATC
AAGCGCAAGGACTTCCGCTGGAAGGACGCCAGCGGGCCCAAGGAGAAGCTGGAGATCTACAAGCCCACTGCCCGGTACT
GCATCCGCTCCATGCTGTCCCTGGAGAGCACCACGCTGGCGGCACAGCACTGCTGCTACGGCGACAACATGCAGCTCAT
CACCAGGGGCAAGGGGGCGGGCACGCCCAACCTCATCAGCACCGAGTTCTCCGCGGAGCTCCACTACAAGGTGGACGTC
CTGCCCTGGATTATCTGCAAGGGTGACTGGAGCAGGTATAACGAGGCCGGCCTCCCAACAACGGACAGAAGTGCACAG
AGAGCCCCTCGGACGAGGACTACATCAAGCAGTTCCAAGAGGCCAGGGAATATTAA

A disclosed NOV15a polypeptide (SEQ ID NO:90) encoded by SEQ ID NO:89 is 539 amino acid residues and is presented using the one-letter amino acid code in Table 15B. SignalP, Psort and/or Hydropathy results predict that NOV15a does not contain a known signal peptide and is likely to be localized to the mitochondrial matrix space with a certainty of 0.6574. In alternative embodiments, NOV151 is localized to the mitochondrial inner membrane with a certainty of 0.3502; the mitochondrial intermembrane space with a certainty of 0.3502; or the mitochondrial outer membrane with a certainty of 0.3502. NOV15a has a molecular weight of 61683.6 Daltons.

Table 15B. Encoded NOV15a protein sequence (SEQ ID NO:90).

GSCCRLRYCRTCSPETSFSLSKEAPREHLDHQAAHQPFPRPRFRQETGHPSLQRDFPRSFLLDLPNFPDLSKADINGQNP
NIQVTIEVVDGPDSEADKDQHPENKPSWSVPSPDWRAWWQRSLSLARANSGDQDYKYDSTSDDSNFLNPRPRGWDHTAPGH
RTFETKDQPEYDSTDGEGDWSLWSVCSVTCGNGNQKRTRSCGYACTATESRTCDRPNCPACTGFLIVKEAWLGVVVWHVP
APPTGNPSVPLPEVFLWTRAQLRMNAQGIPSWKSRTSPLSVMNGSWWIKTQIPINKNKSGLSKERIYSKDYCREARDVIS
LLLQWDERCDHKICKHLKEQPGVTCSLKHLLWAGCTRGERVSLWPFPDTDSCERWMSFKARFLKKYMHKVMNDLPSCPCS
YPTEVAYSTADIFDRIKRKDFRWKDASGPKEKLEIYKPTARYCIRSMLSLESTTLAAQHCCYGDNMQLITRGKGAGTPNL
ISTEFSAELHYKVDVLPWIICKGDWSRYNEARPPNNGQKCTESPSDEDYIKQFQEAREY

NOV15b

A disclosed NOV15b nucleic acid of 1238 nucleotides (also referred to as CG57356-01) encoding a novel novel intracellular thrombospondin domain containing protein-like protein is

shown in Table 15C. A partial open reading frame was identified beginning with an ACG codon at nucleotides 3-5 and ending with a TAA codon at nucleotides 1236-1238. A partial codon upstream from the first in-frame coding triplet is italicized in Table 15C, and the start and stop codons are in bold letters. In further embodiments, the NOV15 coding region extends 5' to the sequence disclosed in Table 15C.

Table 15C. NOV15b Nucleotide Sequence (SEQ ID NO:91)

*GT***ACG**TGTAGTCCTGAAACCAGCTTTTCTCTCTCCAAAGAAGCACCAAGGGAGCATCTGGACCACCAGGCTGCACACCA
ACCCTTCCCCAGACCGCGATTCCGACAAGAGACGGGGCACCCTTCATTGCAAAGAGATTTCCCCAGATCCTTTCTCCTT
GATCTACCAAACTTTCCAGATCTTTCCAAAGCTGATATCAATGGGCAGAATCCAAATATCCAGGTCACCATAGAGGTGG
TCGACGGTCCTGACTCTGAAGCAGATAAAGATCAGCATCCGGAGAATAAGCCCAGCTGGTCAGTCCCATCCCCCGACTG
GCGGGCCTGGTGGCAGAGGTCCCTGTCCTTGGCCAGGGCAAACAGCGGGGACCAGGACTACAAGTACGACAGTACCTCA
GACGACAGCAACTTCCTCAACCCCCCCAGGGGGTGGGACCATACAGCCCCAGGCCACCGGACTTTTGAAACCAAAGATC
AGCCAGAATATGATTCCACAGATGGCGAGGGTGACTGGAGTCTCTGGTCTGTCTGCAGCGTCACCTGCGGGAACGGCAA
CCAGAAACGGACCCGGTCTTGTGGCTACGCGTGCACTGCAACAGAATCGAGGACCTGTGACCGTCCAAACTGCCCAGGA
ATTGAAGACACTTTTAGGACAGCTGCCACCGAAGTGAGTCTGCTTGCGGGAAGCGAGGAGTTTAATGCCACCAAACTGT
TTGAAGTTGACACAGACAGCTGTGAGCGCTGGATGAGCTGCAAAAGCGAGTTCTTAAAGAAGTACATGCACAAGGTGAT
GAATGACCTGCCCAGCTGCCCCTGCTCCTACCCCACTGAGGTGGCCTACAGCACGGCTGACATCTTCGACCGCATCAAG
CGCAAGGACTTCCGCTGGAAGGAGCGCAGCGGGCCCAAGGAGAAGCTGGAGATCTACAAGCCCACTGCCCGGTACTGCA
TCCGCTCCATGCTGTCCCTGGAGAGCACCAGGTGCGGCACAGCACTGCTGCTACGGCGACAACATGCAGCTCATCAC
CAGGGCGAAGGGGCGGGCACGCCCAACCTCATCGGCACCGAGTTCTCCGCGGAGCTCCACTACAAGGTGGACGTCCTG
CCCTGGATTATCTGCAAGGGTGACTGGAGCAGGTATAACGAGGCCCGGCCTCCCAACAACGGACAGGAGTGCACAGAGA
GCCCCTCGGACGAGGACTACATCAAGCAGTTCCAAGAGGCCAGGGAATATT**AA**

A disclosed NOV15b polypeptide (SEQ ID NO:92) encoded by SEQ ID NO:91 is 411 amino acid residues and is presented using the one-letter amino acid code in Table 15D. NOV15b is believed to be a mature protein. SignalP, Psort and/or Hydropathy results predict that NOV15b does not contain a known signal peptide and is likely to be localized in the cytoplasm with a certainty of 0.4500. In alternative embodiments, NOV15b is localized to a microbody (peroxisome) with a certainty of 0.1163; the mitochondrial matrix space with a certainty of 0.1000; or a lysosome (lumen) with a certainty of 0.1000. NOV15b has a molecular weight of 46743.0 Daltons.

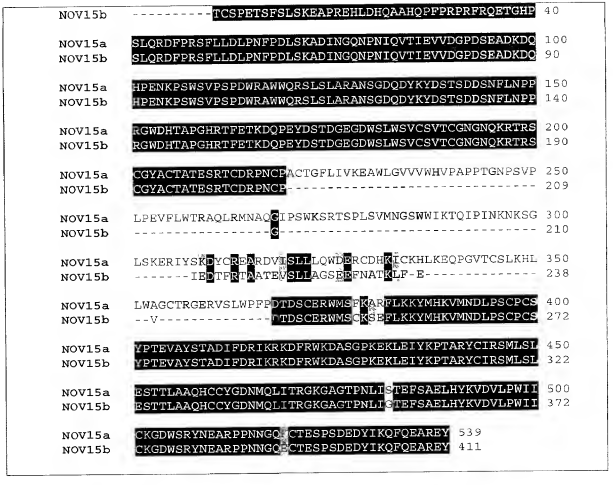
Table 15D. Encoded NOV15b protein sequence (SEQ ID NO:92).

TCSPETSFSLSKEAPREHLDHQAAHQPFPRPRFRQETGHPSLQRDFPRSFLLDLPNFPDLSKADINGQNPNIQVTIEVVDGP
DSEADKDQHPENKPSWSVPSPDWRAWWQRSLSLARANSGDQDYKYDSTSDDSNFLNPPRGWDHTAPGHRTFETKDQPEYDST
DGEGDWSLWSVCSVTCGNGNQKRTRSCGYACTATESRTCDRPNCPGIEDTFRTAATEVSLLAGSEEFNATKLFEVDTDSCER
WMSCKSEFLKKYMHKVMNDLPSCPCSYPTEVAYSTADIFDRIKRKDFRWKDASGPKEKLEIYKPTARYCTRSMLSLESTTLA
AQHCCYGDNMQLITRGKGAGTPNLIGTEFSAELHYKVDVLPWIICKGDWSRYNEARPPNNGQECTESPSDEDYIKQFQEARE
Y

NOV15a and NOV15b are related to each other as shown in the alignment listed in Table 15E.

Table 15E: ClustalW of NOV15 Variants

NOV15a	GSCCRLRYCR	TCSPETSFSLSKEAPREHLDHQAAHQPFPRPRFRQETGHP 50



The novel intracellular thrombospondin domain containing protein-like NOV15 gene maps to chromosome 7. This assignment was made using mapping information associated with genomic clones, public genes and ESTs sharing sequence identity with the disclosed sequence and CuraGen Corporation's Electronic Northern bioinformatic tool. Exons were predicted by homology and the intron/exon boundaries were determined using standard genetic rules. Exons were further selected and refined by means of similarity determination using multiple BLAST (for example, tBlastN, BlastX, and BlastN) searches, and, in some instances, GeneScan and Grail. Expressed sequences from both public and proprietary databases were also added when available to further define and complete the gene sequence. The DNA sequence was then manually corrected for apparent inconsistencies thereby obtaining the sequences encoding the full-length protein.

In a search of sequence databases, it was found, for example. that the NOV15b nucleic acid sequence of this invention has 373 of 512 bases (72%) identical to a gb:GENBANK-ID:AF111168|acc:AF111168.2 mRNA from Homo sapiens (Homo sapiens serine palmitoyl transferase, subunit II gene, complete cds; and unknown genes). The full NOV15b amino acid sequence was found to have 162 of 164 amino acid residues (98%) identical to, and 163 of 164

amino acid residues (99%) similar to, the 361 amino acid residue ptnr:TREMBLNEW-ACC:CAC16127 protein from Homo sapiens (Human) (BA149I18.1 (NOVEL PROTEIN).

The disclosed NOV15a was found to have homology to the amino acid sequences shown in the BLASTP data listed in Table 15F.

Table 15F. BLAST results for NOV15a

Gene Index/ Identifier	Protein/ Organism	Length (aa)	Identity (%)	Positives (%)	Expect
Q9H599; AL133463; CAC16127.2	BA149I18.1 (NOVEL PROTEIN) (FRAGMENT) homo sapiens. 6/2001	391	189/189, (100%)	189/189, (100%)	1e-117
O95432; AF111168; AAD09622.1	HYPOTHETICAL 72.5 KDA PROTEIN. homo sapiens. 6/2001	658	102/172 (59%)	138/172, (80%)	2e-63
Q9BQL4; AL050320; CAC36074.1	DJ1077I2.1 (NOVEL PROTEIN) (FRAGMENT). homo sapiens. 6/2001	60	49/49 (100%)	49/49, (100%)	3e-22
Q23832; U42213; AAC48313.1	MICRONEMAL TRAP-C1 PROTEIN HOMOLOG (FRAGMENT). cryptosporidium wrairi. 6/2001	660	27/61 (44%)	33/61, (54%)	2e-05
TSP1_HUMAN; P07996; M25631; AAA36741; CAA28370; CAA32889; AAA61178; AAB59366	THROMBOSPONDIN 1 PRECURSOR. homo sapiens. 10/1996	1170	24/54 (44%)	31/54, (57%)	3e-05

The disclosed NOV15b was found to have homology to the amino acid sequences shown in the BLASTP data listed in Table 15G.

Table 15G. BLAST results for NOV15b

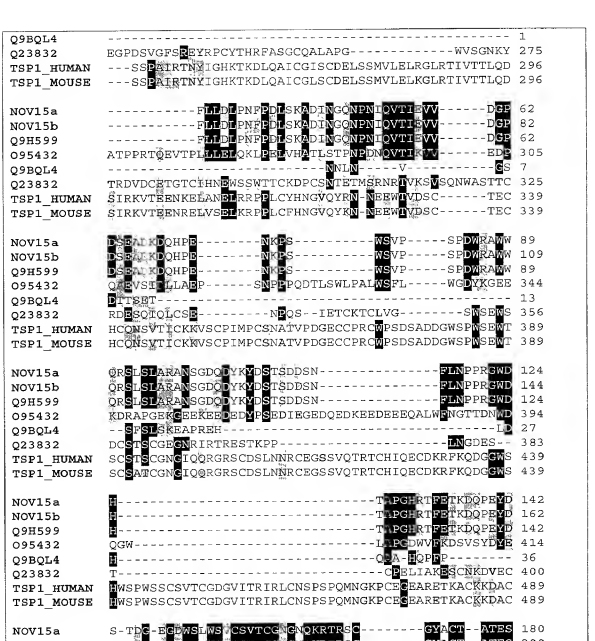
Gene Index/ Identifier	Protein/ Organism	Length (aa)	Identity (%)	Positives (%)	Expect
Q9H599; AL133463; CAC16127.2	BA149I18.1 (NOVEL PROTEIN) (FRAGMENT). homo sapiens. 6/2001	391	390/391, (100%)	390/391, (100%)	0.0
O95432; AF111168; AAD09622.1	HYPOTHETICAL 72.5 KDA PROTEIN. homo sapiens. 6/2001	658	183/392 (47%)	242/392, (62%)	2e-95
Q9BQL4; AL050320; CAC36074.1	DJ1077I2.1 (NOVEL PROTEIN) (FRAGMENT). homo sapiens. 6/2001	60	49/49 (100%)	49/49, (100%)	2e-22
TSP1_HUMAN; P07996; M25631; AAA36741; CAA28370; CAA32889; AAA61178; AAB59366	THROMBOSPONDIN 1 PRECURSOR. homo sapiens. 10/1996	1170	24/54 (44%)	31/54, (57%)	2e-05
TSP1_MOUSE; P35441; AAA50611; AAA40431	THROMBOSPONDIN 1 PRECURSOR. mus musculus. 10/1996	1170	23/54 (43%)	31/54, (57%)	4e-05

The homology of these sequences is shown graphically in the ClustalW analysis shown in Table 15H.

Table 15H Information for the ClustalW proteins

1) NOV15a (SEQ ID NO:90)
2) NOV15b (SEQ ID NO:92)
3) Q9H599 (SEQ ID NO:93)
4) O95432 (SEQ ID NO:94)
5) Q9BQL4 (SEQ ID NO:95)
6) Q23832 (SEQ ID NO:96)
7) TSP1_HUMAN N-ter fragment (SEQ ID NO:97)
8) TSP1_MOUSE N-ter fragment (SEQ ID NO:98)

```
NOV15a      ------------------------------------------------------- 1
NOV15b      ------------------------------------------------------- 1
Q9H599      ------------------------------------------------------- 1
O95432      -MRALRDRAGLLLCVLLLAALLEAALG------LPVKKPRLRGPRPGSLT 43
Q9BQL4      ------------------------------------------------------- 1
Q23832      KLTHYSVGGHASTSRVKGRSSSGSSSG--------D--FKVPGLNG-YLC 39
TSP1_HUMAN  -MGLAWGLGVLFLMHVCGTNRIPESGGDNSVFDIFELTGAARKGSGRRLV 49
TSP1_MOUSE  -MLLRGLGVLFLLHMCGSNRIPESGGDNGVFDIFELIGGARRGPGRRLV 49

NOV15a      ------------------------------------------------------- 1
NOV15b      ------------------------------------------------------- 1
Q9H599      ------------------------------------------------------- 1
O95432      RLAEVSGGGTGLRSALSVPPPQPAGSSRAGSGTGTHT-----GSDPPMER 88
Q9BQL4      ------------------------------------------------------- 1
Q23832      PSYNRDPRGFGCFGLNTAYTVKKNSWQECANQCYWSKVTIYGNCQRSVYN 89
TSP1_HUMAN  KGPDPSSPAFRIEDANLIPPVPDDKFQDLVDAVRTEKGFLLLASLRQMKK 99
TSP1_MOUSE  KGQDLSSPAFRIENANLIPAVPDDKFQDLLDAVWADKGFIFLASLRQMKK 99

NOV15a      ------------------------------------------------------- 1
NOV15b      ------------------------------------------------------- 1
Q9H599      ------------------------------------------------------- 1
O95432      GAGAGRKLPDTGRCPVTEGSTVQLIAPWNAADVHSHGDKDSQTCIRVSAS 138
Q9BQL4      ------------------------------------------------------- 1
Q23832      SNNQDCHLKGGDNDCMKSPDGMILTNRQSYMIGECATTCTVSSWSSWTPC 139
TSP1_HUMAN  TRGTLLAIERKDHSGQVFSVVSNGKAGTLDLSLTVQGKQHVVSVEEALLA 149
TSP1_MOUSE  TRGTLLAMERKDNTGQIFSVVSNGKAGTLDLSLSLPGKQQVVSVEEALLA 149

NOV15a      ------------------------------------------------------- 1
NOV15b      ------------------------------------------------------- 1
Q9H599      ------------------------------------------------------- 1
O95432      PDPRPLKEEEEAPLLPRTHLQAEPHQHGCWTVTEPAAMTPGNATPPRT-- 186
Q9BQL4      ------------------------------------------------------- 1
Q23832      SGVCGEMRSRTRSVLSFPRYDHEYCP-HLIEYSNCVVQNKCPENCPQYGV 188
TSP1_HUMAN  TGQWKSITLFVQEDRAQLYIDCEKMENAELDVPIQSVFTRDLASIARLRI 199
TSP1_MOUSE  TGQWKSITLFVQEDRAQLYIDCDKMESAELDVPIQSIFTRDLASVARLRV 199

NOV15a      ----------------------------HQAAHQGFPREFRQETG--- 18
NOV15b      ---------TCSPETSFSLSKEAPREHLDHQAAHQGFPREFRQETG--- 38
Q9H599      ----------------------------HQAAHQGFPREFRQETG--- 18
O95432      --PEVTPLRLELQKLPGLANTTLSTPNPDTQASASGDPREPREERARLL 234
Q9BQL4      ------------------------------------------------------- 1
Q23832      SILGWGCQFESMFSPNKNLFVSYEEDWKGCMSTCKQDEPCVAWSYNATLS 238
TSP1_HUMAN  AKGGVNDNFQGVLQNVRFVFGTTPEDILRNKGCSSSTSVLLTLDNNVVNG 249
TSP1_MOUSE  AKGDVNDNFQGVLQNVRFVFGTTPEDILRNKGCSSSTNVLLTLDNNVVNG 249

NOV15a      ----HPSLQRDPPRS---------------------------------- 29
NOV15b      ----HPSLQRDPPRS---------------------------------- 49
Q9H599      ----HPSLQRDPPRS---------------------------------- 29
O95432      ---PRTHLQAELHQHGCWTVTEPA----------------ALTPGN 261
```



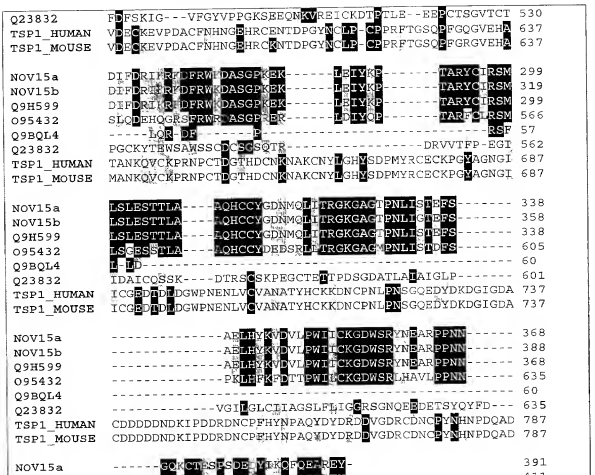


Table 15I lists the domain description from DOMAIN analysis results against NOV15a, and in the analogous regions for NOV15b. This indicates that the NOV15a sequence has properties similar to those of other proteins known to contain this domain.

Table 15I. Domain Analysis of NOV15a

PFAM HMM Domain Analysis of NOV15

Model	Description	Score	E-value
tsp_1 (InterPro)	Thrombospondin type 1 domain	32.5	9.8e-06

Parsed for domains:

Model	Domain	seq-f	seq-t	hmm-f	hmm-t	score	E-value
tsp_1	1/1	178	218 ..	1	54 []	32.5	9.8e-06

Smallest Sum

ProDom Sequences producing High-scoring Segment Pairs:			High Score	Probability P(N)
prdm:1719	p36	(14) FSPO(5) TSP1(3) TSP2(2) - PRECURSOR...	110	3.0e-06
prdm:873	p36	(25) TSP1(9) TSP2(4) PROP(3) - COMPLEMEN...	91	0.00033
prdm:36045	p36	(1) SSP2_PLAYO - SPOROZOITE SURFACE PROTE...	85	0.0014
prdm:1268	p36	(18) CSP(18) - CIRCUMSPOROZOITE PROTEIN ...	74	0.022
prdm:53698	p36	(1) FSPO_XENLA - F-SPONDIN PRECURSOR. GLY...	62	0.35

BLOCKS Protein Domain Analysis

AC#		Description	Strength	Score
BL00612B	0	Osteonectin domain proteins.	1891	1066
BL00652C	0	TNFR/NGFR family cysteine-rich region protein	1217	1062
BL00979I	0	G-protein coupled receptors family 3 proteins	1459	1059
BL00641E	0	Respiratory-chain NADH dehydrogenase 75 Kd su	1700	1039
BL00512A	0	Alpha-galactosidase proteins.	1403	1035
BL00096G	0	Serine hydroxymethyltransferase pyridoxal-pho	1543	1030

The thrombospondin repeat was first described in 1986 by Lawler & Hynes. It was found in the thrombospondin protein where it is repeated 3 times. Now a number of proteins involved in the complement pathway (properdin. C6, C7, C8A, C8B, C9) as well as extracellular matrix protein like mindin, F-spondin, SCO-spondin and even the circumsporozoite surface protein 2 and TRAP proteins of Plasmodium have been shown to contain one or more instances of this repeat. It has been involved in cell-cell interaction, inhibition of angiogenesis, and apoptosis.

The intron-exon organisation of the properdin gene confirms the hypothesis that the repeat might have evolved by a process involving exon shuffling. A study of properdin structure provides some information about the structure of the thrombospondin type I repeat.

BLASTP analysis shows that NOV15 has 24 of 55 (43%) identical to, and 27 of 55 (49%) positive with, the 57 aa p36 (14) FSPO(5) TSP1(3) TSP2(2) - precursor glycoprotein signal repeat cell adhesion EGF-like domain thrombospondin calcium binding (prdm:1719, Expect = 3.0e-06); 15 of 35 (42%) identical to, and 18 of 35 (51%) positive with, the 54 aa p36 (25) TSP1(9) TSP2(4) PROP(3) - complement precursor repeat signal glycoprotein EGF-like domain pathway thrombospondin cell (prdm:873, Expect = 0.00033); 20 of 68 (29%) identical to, and 28 of 68 (41%) positive with, the 108 aa p36 (1) SSP2_PLAYO - sporozoite surface protein 2 precursor, malaria; sporozoite; repeat; signal; antigen; transmembrane (prdm:36045, Expect = 0.0014); 23 of 59 (38%) identical to, and 28 of 59 (47%) positive with, the 87 aa p36 (18) CSP(18) - circumsporozoite protein precursor CS malaria sporozoite repeat signal (prdm:1268, Expect = 0.022); and 10 of 21 (47%) identical to, and 13 of 21 (61%) positive with, the 59 aa p36 (1) FSPO_XENLA - F-spondin precursor, glycoprotein; signal; repeat; cell adhesion (prdm:53698, Expect = 0.43).

PROSITE analysis of NOV15a shows that the NOV15a polypeptide has two N-glycosylation sites (Pattern-ID: ASN_glycosylation PS00001 (Interpro)); four Protein kinase C phosphorylation sites (Pattern-ID: PKC_PHOSPHO_SITE PS00005 (Interpro)); eight Casein

kinase II phosphorylation sites (Pattern-ID: CK2_PHOSPHO_SITE PS00006 (Interpro)); one Tyrosine kinase phosphorylation site (Pattern-ID: TYR_PHOSPHO_SITE PS00007 (Interpro)); and four N-myristoylation sites (Pattern-ID: MYRISTYL PS00008 (Interpro)). PROSITE analysis of NOV15b shows that the NOV15b polypeptide has one N-glycosylation site (Pattern-ID: ASN_ glycosylation PS00001 (Interpro)); three Protein kinase C phosphorylation sites (Pattern-ID: PKC_PHOSPHO_SITE PS00005 (Interpro)); seven Casein kinase II phosphorylation sites (Pattern-ID: CK2_PHOSPHO_SITE PS00006 (Interpro)); one Tyrosine kinase phosphorylation site (Pattern-ID: TYR_PHOSPHO_SITE PS00007 (Interpro)); and four N-myristoylation sites (Pattern-ID: MYRISTYL PS00008 (Interpro)).

In a BlastP analysis of a public database, NOV15a was found to have 185 of 188 aa residues aa residues (98%) identical to, and 188 of 188 aa residues (100%) positive with, the 198 aa Human ORFX ORF1686 polypeptide sequence SEQ ID NO:3372 (patp:AAB41922, Expect = 7.8e-106) (NOV15b has 185/188 aa (98%) identical, 188/188 aa (100%) positive). NOV15a has 102 of 172 aa residues (59%) identical to, and 138 of 172 aa residues (80%) positive with, the 571 aa Human proliferation differentiation factor amino acid sequence (patp:AAB49765, Expect = 1.2e-90) (NOV15b has 155/290 aa (53%) identical, 205/290aa (70%) positive). NOV15a has 102 of 172 aa residues (59%) identical to, and 138 of 172 aa residues (80%) positive with, the 571 aa Human membrane or secretory protein clone PSEC0137 (patp:AAB88393, Expect = 1.2e-90) (NOV15b has 155/290 aa (53%) identical, 205/290 aa (70%) positive). NOV15a has 24 of 54 aa residues (44%) identical to, and 31 of 54 aa residues (57%) positive with, the 57 aa Human METH1 thombospondin-like domain #3 (patp:AAY49505, Expect = 3.2e-06) (NOV15b has 24/54 aa (44%) identical, 31/54 aa (57%) positive). NOV15a has 24 of 54 aa residues (44%) identical to, and 31 of 54 aa residues (57%) positive with, the 57 aa Homo sapiens TSP1 domain (patp:AAB50007, Expect = 3.2e-06) (NOV15b has 24/54 aa (44%) identical, 31/54 aa (57%) positive). The Patp BLAST results for NOV15a and NOV15b are listed in Table 15J.

Table 15J. Patp alignments of NOV15

Sequences producing High-scoring Segment Pairs.	High Score	Smallest P(N) NOV15a	Sum Prob. P(N) NOV15b
patp:AAB41922 Human ORFX ORF1686 polypeptide seque...	1048	7.8e-106	7.8e-106
patp:AAB49765 Human proliferation differentiation ...	616	1.2e-90	5.2e-95
patp:AAB88393 Human membrane or secretory protein ...	616	1.2e-90	5.2e-95
patp:AAY49505 Human METH1 thombospondin-like doma ...	118	3.2e-06	2.1e-06
patp:AAB50007 TSP1 domain #3 - Homo sapiens, 57 aa.	118	3.2e-06	2.1e-06

The homologies shown above are shared by NOV15b insofar as NOV15b is homologous to NOV15a as shown in Table 15E.

The novel intracellular thrombospondin domain containing protein-like NOV15 gene disclosed in this invention is expressed in at least the following tissues: lung, testis, b-cell. Expression information was derived from the tissue sources of the sequences that were included in the derivation of the sequence, as described in Example 1.

The above defined information for this invention suggests that these novel intracellular thrombospondin domain containing protein-like NOV15 proteins may function as a member of a "novel intracellular thrombospondin domain containing protein-like family". Therefore, the NOV15 nucleic acids and proteins identified here may be useful in potential therapeutic applications implicated in (but not limited to) various pathologies and disorders as indicated below.

The protein similarity information, expression pattern, cellular localization, and map location for the protein and nucleic acid disclosed herein suggest that this novel intracellular thrombospondin domain containing protein-like NOV15 protein may have important structural and/or physiological functions characteristic of the novel intracellular thrombospondin domain containing protein family. Therefore, the NOV15 nucleic acids and proteins are useful in potential diagnostic and therapeutic applications and as a research tool. These include serving as a specific or selective nucleic acid or protein diagnostic and/or prognostic marker, wherein the presence or amount of the nucleic acid or the protein are to be assessed. These also include potential therapeutic applications such as the following: (i) a protein therapeutic, (ii) a small molecule drug target, (iii) an antibody target (therapeutic, diagnostic, drug targeting/cytotoxic antibody), (iv) a nucleic acid useful in gene therapy (gene delivery/gene ablation), (v) an agent promoting tissue regeneration *in vitro* and *in vivo*, and (vi) a biological defense weapon.

The NOV15 nucleic acids and proteins have applications in the diagnosis and/or treatment of various diseases and disorders. For example, the compositions of the present invention will have efficacy for the treatment of patients suffering from: systemic lupus erythematosus, autoimmune disease, asthma, emphysema, scleroderma, allergy, ARDS; fertility, hypogonadism; immunological disease and disorders as well as other diseases, disorders and conditions.

Based on the tissues in which NOV15 is most highly expressed; including Thryoid, heart, uterus, mammary gland, pituitary gland, lymph node, placenta, brain, pancreas, and spleen; specific uses include developing products for the diagnosis or treatment of a variety of diseases and disorders. Additional disease indications and tissue expression for NOV15 is presented in Example 2.

NOV15 nucleic acids and polypeptides are further useful in the generation of antibodies that bind immuno-specifically to the novel NOV15 substances for use in therapeutic or

diagnostic methods. These antibodies may be generated according to methods known in the art, using prediction from hydrophobicity charts, as described in the "Anti-NOVX Antibodies" section below. For example the disclosed NOV15 proteins have multiple hydrophilic regions, each of which can be used as an immunogen. In one embodiment, a contemplated NOV15a epitope is from about amino acids 1 to 70. In additional embodiments, NOV15a epitopes are from about amino acids 175 to 230 and from about amino acids 250 to 539. In another embodiment, a NOV15b epitope is from about amino acids 1 to 60. In further embodiments, NOV15b epitopes are from about amino acids 65 to 225, from about amino acids 230 to 320 and from about amino acids 325 to 411. This novel protein also has value in development of powerful assay system for functional analysis of various human disorders, which will help in understanding of pathology of the disease and development of new drug targets for various disorders.

NOV16

NOV16 includes two novel FYVE finger-containing phosphoinositide kinase-like proteins disclosed below. The disclosed proteins have been named NOV16a and NOV16b.

NOV16a

A disclosed NOV16a nucleic acid of 2760 nucleotides (also referred to as 101330077 and 100391903) encoding a novel FYVE-finger kinase/Transposase-like protein is shown in Table 16A. An open reading frame was identified beginning with an ATG initiation codon at nucleotides 898-900 to and ending with a TGA codon at nucleotides 1516-1518. A putative untranslated region upstream from the initiation codon and downstream from the termination codon is underlined in Table 16A, and the start and stop codons are in bold letters.

Table 16A. NOV16a Nucleotide Sequence (SEQ ID NO:99)

CCGGGGGCGCAGCCGCGGGCCCACCTCGGCCTCCCCTGAGCGGACGCCTCCCGCGCGCACCGGGGGCCCCGGAGACCG
CCTTCCCCGCTCCGAACGCACGCGGCCCGGCCCCGGCCGAGGTGCCTGAACGCTACCCGAGCTGCGGCGGGGCTCCCGGG
GTGAGTGCTGCAGCCCCAGGCCCGCCTGCTCCCACAGGCTCGGGCAATGGAGACCCGCGGCCGCCCCGCCCCTTGACC
CTGCCTCACCCCTCACGCCCGCTGCCGCCCACGACCTCCGACCCGCTGCCGCCCGGCTCGCAGCCCGGCTCGCAGCCC
GGCTCGGCGGGCCTCACCTCCCGCGGGTTCCGCACTCCTCTCCCGCCGTCCTGCTCCTCGGCCTTCCCTCCAATA
GGCGCCTAGCACCCTGAGTGGGCTACACCAATCAGAGACGAAGCGGCGCTAACGTGACTGACTAACTAACCAATCCAAA
GTCTCAATCTCCCTGAGAGGGGCGGAGCGTACCCGGGCCAGCCCTCCGCCGCCGATTGGTGATCGACCTCAGGGTTGCAG
GGGCGGTGCCCTTACACGGATTGGAGAGGGCAGCGATGGGGCGGAGTTCAAGCTCCGATTAGTCCGCGCTCCGTGGCGG
GCTTGGCGATTGGACGCCGGCGCTGTCAGCCGCGGCGGGACCGGGGCGGGGCGGGCGGTGCCCCGGGCTGGGCGAGGGG
CCGGGTGCGGGGCCGCTGGCCGAGAGGCTGAGGCGGCGTCATGTCCTCCGAGGTGTCCGCGCGCCGCGACGCCAAGAAG
CTGGTGCGCTCCCCGAGCGGCCTGCGCATGGTGCCCGAACACCGCGCCTTCGGAAGCCCGTTCGGCCTGGAGGAGCCGC
AGTGGGTCCCGGACAAGGAGGTGGGTGTATGCAGTGTGACGCCAAGTTTGACTTTCTCACCAGAAAGCACCACTGTCGC
CGCTGCGGGAAGTGCTTCTGCGACAGGTGCTGCAGCCAGAAGGTGCCGCTGCGGCGCATGTGCTTGTGGACCCCGTGC
GGCAGTGCGCGGAGTGCGCCCTGGTGTCCCTCAAGGAGGCGGAGTTCTACGACAAGCAGCTCAAAGTGCTCCTGAGCGG
AGCCACCTTCCTCGTCACGTTTGGAAACTCAGAGAAACCTGAAACTATGACTTGTCGTCTTTCCAATAACCAGAGATAC
TTGTTTCTGGATGGAGACAGCCACTATGAAATCGAAATTGTACACATTTCCACCGTGCAGATCCTCACAGAAGGCTTCC
CTCCTGGAGAAAAAGACATTCACGCTTACACCAGCCTCCGGGGGAGCCAGCCTGCCTCTGAAGGAGGCAACGCACGGGC
CACAGGCATGTTCCTGCAGTATACAGTGCCGGGGACGGAGGGTGTGACCCAGCTGAAGCTGACAGTGGTGGAGGACGTG
ACTGTGGGCAGGAGGCAGGCGGTGGCGTGGCTAGTGATCTGCAGGCTGCCAAGCTCCTCTATGAATCTCGGGACCAGTA
ACTCTACGTGGGGCTGAGCTTGGAGTACGTGTGGTCACCAGGACTGAGTCGCTTGGAACAGCAGAGCCTGCTCCTTGCG

TACCACAGGGATTAATCCTGCTTGTGCTGGGAAATGCAACTCACTCATGTATTTGGAGAAACAGGAGTGTTCACTTATC
TAGTGCAATATGTTCACAGTTTATTAATGCTTTAAACAGCTTCATGTTTTAGAATTTGTGTATTGTCAATACTTAATTG
GGGGTGGGAGAGACTGAGCTACACTACTGCTAAACTATTTTAGCATAATATATACCATTTTATGAGTTCGCAGGTCT
ACTAGAAGGTTCTGGCCCATCAATATTCATTTCATTTAATTCTTCCACAGAACCAGTTTGGGCAGTAGGAACTCAGGCT
TCTGGTCTGCAGTGGAGCCTGTTCGCCTCTAATAGCCAGTTTACAGCACTTGCCTTAGCCTGTTTCACAGACTTGTCCA
CTTACCTTGTCACTAATTTGGGGCTTCTGGGCTGTGAGTGATCCTTGATACTTCACCAAGGGGAACGTGGGGGCTTTG
TGTTTTGTACTTTTCACTCACTATTTCACTTTATTAAGATGACTGTACAGCAATTTGTATATAAAGCTATGATTAAAA
ACTATTTTGAACATACGGACAAGGCCTCGCCTTCCTGTGTCCAGATCACCTGAACCCTCGTGCCACAGCGCAGTCTGGG
TCCAGAAAGAAGACTCACAGCCGCCGGGGTGAGACGGGTTTATTGTGCACATTTACACAGCGTCAGCAGCGTCTGGGCT
GGCAGCGGCCATGCTCCTGTGGTCGGGCTGCTCTACAAGGGCGTTCACTTTTCTTCACCACACTATGTACAGTCAGTGC
TCCAAGGTGATGGGCTACAGTGCTGCATCAGTGAGTCTGTACACACATTTTTACATAAATTACACACGACTCATACATG
AAAAATAGAGCCTAAGGGCCTGTATTTTAATGAGAAAAAAAAATTTCCAACATAGTTCGGGTAGCTTTGAATGGTCTA
GTCAAAAAATACTTTGTATATAAAAGCCTGTACGTACAATTCACACCTCAGTGAAGCGCCCTCCTTGCCTTGAGGC
TGGGCCTGGACAAAGGTGGCCTCACAGCCAGCCCAGGCAGGGAGATCGGCAGAGAGGGGTGGCCCCTGACCCCAGCTC
CTCTGCCCCAGCTGCTGCTCCTTGGTGGCGGCCCCTCCTGACACCAGGCGTCTGCCATCCTTCAGGCACCAAAC

A disclosed NOV16a polypeptide (SEQ ID NO:100) encoded by SEQ ID NO:99 is 206 amino acid residues and is presented using the one-letter amino acid code in Table 16B. SignalP, Psort and/or Hydropathy results predict that NOV16b has no known signal peptide and is likely to be localized in the cytoplasm with a certainty of 0.6500. In alternative embodiments, NOV16b is localized to the mitochondrial matrix space with a certainty of 0.1000, lysosome (lumen) with a certainty of 0.1000, or perhaps the endoplasmic reticulum (membrane) with a certainty of < 0.0001. NOV16a has a molecular weight of 23030.2 Daltons.

Table 16B. Encoded NOV16a protein sequence (SEQ ID NO:100).

MQCDAKFDFLTRKHHCRRCGKCFCDRCCSQKVPLRRMCFVDPVRQCAECALVSLKEAEFYDKQLKVLLSGATFLV
TPGNSEKPETMTCRLSNNQRYLFLDGDSHYEIEIVHISTVQILTEGFPPGEKDIHAYTSLRGSQPASEGGNARAT
GMFLQYTVPGTEGVTQLKLTVVEDVTVGRRQAVAWLVICRLPSSSMNLGTSNSTWG

NOV16b

A disclosed NOV16b nucleic acid of 673 nucleotides (also referred to as CG57248-01) encoding a novel FYVE-finger kinase/Transposase-like protein is shown in Table 16C. An open reading frame was identified beginning with an ATG initiation codon at nucleotides 44-46 and ending with a TAA codon at nucleotides 650-652. A putative untranslated region upstream from the initiation codon and downstream from the termination codon is underlined in Table 16C, and the start and stop codons are in bold letters.

Table 16C. NOV16b Nucleotide Sequence (SEQ ID NO:101)

GTTCCAACTATTTTGTCCGCCCACAGGAATTCGCCCTTGGTGT**ATG**CAGTGTGACGCCAAGTTTGACTTTCTCACCA
GAAAGCACCACTGTCGCCGCTGCGGGAAGTGCTTCTGCGACAGGTGCTGCAGCCAGAAGGTGCCGCTGCGGCGCATG
TGCTTTGTGGACCCCGTGCGGCAGTGCGCGGAGTGCGCCCTGGTGTCCCTCAAGGAGGCGGAGTTCTACGACAAGCA
GCTCAAAGTGCTCCTGAGCGGAGCCACCTTCCTCGTCACGTTTGGAAACTCAGAGAAACCTGAAACTATGACTTGTC
GTCTTTCCAATAACCAGAGATACTTGTTTCTGGATGGAGACAGCCACTATGAAATCGAAATTGTACACATTTCCACC
GTGCAGATCCTCACAGAAGGCTTCCCTCCTGGAGAAAAAGACATTCACGCTTACACCAGCCTCCGGGGGAGCCAGCC
TGCCTCTGAAGGAGGCAACGCACAGGCCACAGGCATGTTCCTGCAGTATACAGTGCCGGGACGGAGGGTGTGACCC
AGCTGAAGCTGACAGTGGTGGAGGACGTGACTGTGGCAGGAGGCAGGCCGTGGCGTGGCTAGTGGCCATGCACAAG
GCTGCCAAGCTCCTCTATGAATCTCGGGACCAG**TAA**CTCTACGTGGGGCTGAGCTTG

A disclosed NOV16b polypeptide (SEQ ID NO:102) encoded by SEQ ID NO:101 is 202 amino acid residues and is presented using the one-letter amino acid code in Table 16D. SignalP, Psort and/or Hydropathy results predict that NOV16b has no known signal peptide and is likely to be localized in the cytoplasm with a certainty of 0.4500. In alternative embodiments, NOV16b is localized to the microbody (peroxisome) with a certainty of 0.3000, a mitochondrial matrix space with a certainty of 0.1000, or a lysosome (lumen) with a certainty of 0.1000. NOV16b has a molecular weight of 22751.9 Daltons.

Table 16D. Encoded NOV16b protein sequence (SEQ ID NO:102).

```
MQCDAKFDFLTRKHHCRRCGKCFCDRCCSQKVPLRRMCFVDPVRQCAECALVSLKEAEFYDKQLKVLLSGATFLV
TFGNSEKPETMTCRLSNNQRYLFLDGDSHYEIEIVHISTVQILTEGFPPGEKDIHAYTSLRGSQPASEGGNAQAT
GMFLQYTVPGTEGVTQLKLTVVEDVTVGRRQAVAWLVAMHKAAKLLYESRDQ
```

The FYVE finger-containing phosphoinositide kinase-like gene disclosed in this invention maps to chromosome 14. This assignment was made using mapping information associated with genomic clones, public genes and ESTs sharing sequence identity with the disclosed sequence and CuraGen Corporation's Electronic Northern bioinformatic tool. NOV16a and NOV16b are related to each other as shown in the alignment listed in Table 16E.

Table 16E: ClustalW of NOV16 Variants

```
16      MQCDAKFDFLTRKHHCRRCGKCFCDRCCSQKVPLRRMCFVDPVRQCAECA 50
16-1    MQCDAKFDFLTRKHHCRRCGKCFCDRCCSQKVPLRRMCFVDPVRQCAECA 50

16      LVSLKEAEFYDKQLKVLLSGATFLVTFGNSEKPETMTCRLSNNQRYLFLD 100
16-1    LVSLKEAEFYDKQLKVLLSGATFLVTFGNSEKPETMTCRLSNNQRYLFLD 100

16      GDSHYEIEIVHISTVQILTEGFPPGEKDIHAYTSLRGSQPASEGGNAQAT 150
16-1    GDSHYEIEIVHISTVQILTEGFPPGEKDIHAYTSLRGSQPASEGGNAQAT 150

16      GMFLQYTVPGTEGVTQLKLTVVEDVTVGRRQAVAWLVICRLPSSSMNLGT 200
16-1    GMFLQYTVPGTEGVTQLKLTVVEDVTVGRRQAVAWLVAMHKAAKLLYESR 200

16      SNSTWG 206
16-1    DQ---- 202
```

The disclosed NOV16a amino acid sequence has homology to the amino acid sequences shown in the BLASTP data listed in Table 16F.

Table 16F. BLAST results for NOV16a

Gene Index/ Identifier	Protein/ Organism	Length (aa)	Identity (%)	Positives (%)	Expect
Q9BQ24; BC005999; AAH05999.1; AAH01130	HYPOTHETICAL 26.5 KDA PROTEIN (UNKNOWN) (PROTEIN FOR MGC:2550). homo sapiens. 6/2001	234	169/187 (90%)	169/187, (90%)	7e-95
Q9D1E2; AK003661; BAB22923.1	1110013H04RIK PROTEIN. mus musculus. 6/2001	212	136/186 (73%)	145/186, (78%)	4e-75

FYV1_MOUSE; Q9Z1T6; AF102777; AAD10191.1	FYVE finger-containing phosphoinositide kinase (EC 2.7.1.68)(1-phosphatidylinositol-4-phosphate kinase) (PIP5K) (PTDINS(4)P-5-KINASE) (P235). mus musculus. 5/2000	2052	35/113 (31%)	56/113, (50%)	3e-09
Q9HCC9; AB046863; BAB13469.1	KIAA1643 PROTEIN (FRAGMENT). homo sapiens. 6/2001	993	26/47 (55%)	27/47, (57%)	5e-09
Q9CVQ1; AK007036; BAB24835.1	1700092A20RIK PROTEIN (FRAGMENT).	173	23/47 (49%)	28/47, (60%)	8e-09

In a search of sequence databases, it was found, for example, that the NOV16 nucleic acid sequence of this invention has 208 of 215 bases (96%) identical to a gb:GenBank-ID:AK001921|acc:AK001921.1 mRNA from Homo sapiens (Homo sapiens cDNA FLJ11059 fis, clone PLACE1004740). The full NOV16 amino acid sequence was found to have 37 of 111 amino acid residues (33%) identical to, and 61 of 111 amino acid residues (54%) similar to, the 2052 amino acid residue ptnr:SWISSNEW-ACC:Q9Z1T6 protein from Mus musculus (Mouse) (FYVE finger-containing phosphoinositide kinase (EC 2.7.1.68) (1-phospatidylinositol-4-phosphate kinase) (PIP5K) (PTDINS(4)P-5-KINASE) (P235)).

The disclosed NOV16b amino acid sequence has homology to the amino acid sequences shown in the BLASTP data listed in Table 16G.

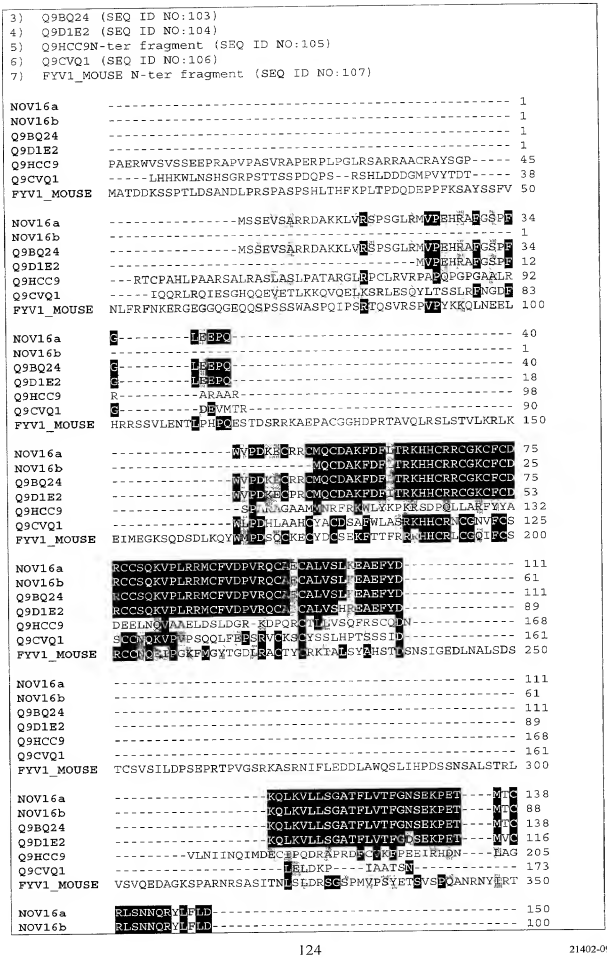
Table 16G. BLAST results for NOV16b

Gene Index/ Identifier	Protein/ Organism	Length (aa)	Identity (%)	Positives (%)	Expect
Q9BQ24	HYPOTHETICAL 26.5 KDA PROTEIN (UNKNOWN) (PROTEIN FOR MGC:2550). homo sapiens. 6/2001	234	183/202 (91%)	184/202, (91%)	1e-103
Q9D1E2	1110013H04RIK PROTEIN. mus musculus. 6/2001	212	150/202 (74%)	159/202, (79%)	2e-83
FYV1_MOUSE	FYVE FINGER-CONTAINING PHOSPHOINOSITIDE KINASE (EC 2.7.1.68)(1-PHOSPHATIDYLINOSITOL-4-PHOSPHATE KINASE) (PIP5K) (PTDINS(4)P-5-KINASE) (P235). mus musculus. 5/2000	2052	35/113 (31%)	56/113, (50%)	3e-09
Q9HCC9	KIAA1643 PROTEIN (FRAGMENT).	993	26/47 (55%)	27/47, (57%)	5e-09
Q9CVQ1	1700092A20RIK PROTEIN (FRAGMENT).	173	23/47 (49%)	28/47, (60%)	8e-09

The homology of these sequences is shown graphically in the ClustalW analysis shown in Table 16H.

Table 16H Information for the ClustalW proteins

1) NOV16a (SEQ ID NO:100)
2) NOV16b (SEQ ID NO:102)


3) Q9BQ24 (SEQ ID NO:103)
4) Q9D1E2 (SEQ ID NO:104)
5) Q9HCC9N-ter fragment (SEQ ID NO:105)
6) Q9CVQ1 (SEQ ID NO:106)
7) FYV1_MOUSE N-ter fragment (SEQ ID NO:107)
NOV16a
NOV16b
Q9BQ24
Q9D1E2
Q9HCC9
Q9CVQ1
FYV1_MOUSE

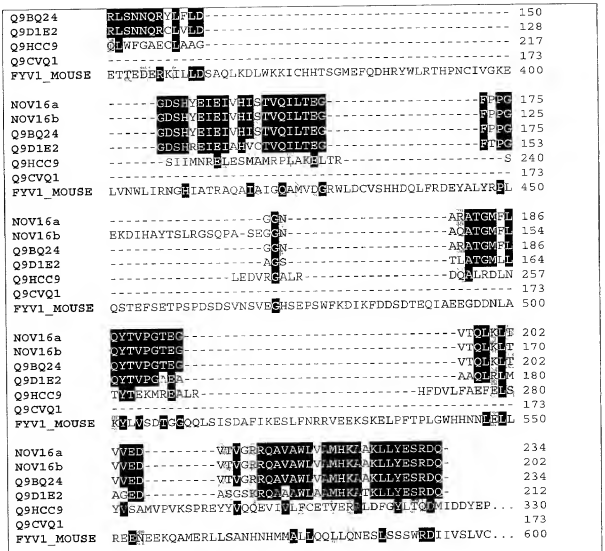


Table 16I lists the domain description from DOMAIN analysis results against NOV16a. This indicates that the NOV16a sequence has properties similar to those of other proteins known to contain this domain.

Table 16I. Domain Analysis of NOV16a

PFAM HMM Domain Analysis of NOV16

Model	Domain	seq-f	seq-t	hmm-f	hmm-t	score	E-value
FYVE	1/1	1	49 [.	13	66 ..	29.1	8.9e-07

PRODOM analysis of Nov16

prdm:3303 p36 (8) FGD1(2) - PROTEIN KINASE RHO/RAC FACTOR ZINC-FINGER PUTATIVE GUANINE NUCLEOTIDE EXCHANGE GEF, 235 aa
Expect = 0.00015, identity= 20/50 (40%), positive=24/50 (48%) for NOV16a: 1 to 49;
Sbjct: 148 to 197

prdm:28902 p36 (1) YLN2_CAEEL - HYPOTHETICAL 46.2 KD TRP-ASP REPEATS CONTAINING PROTEIN D2013.2 IN CHROMOSOME II. HYPOTHETICAL PROTEIN; REPEAT; WD REPEAT, 138 aa
Expect = 0.0019, identity=14/38 (36%), positive=18/38 (47%) for NOV16a: 12 to 49;
Sbjct: 38 to 75

```
prdm:4778 p36 (5) - INHIBITOR SERINE PROTEASE CHYMOTRYPSIN/ELASTASE PROTEIN TRYPSIN
ISOINHIBITOR ISOINHIBITORS R10H1.1 CHROMOSOME, 67 aa
Expect = 0.053, identity=14/36 (38%), positive=21/36 (58%), for NOV16a: 18 to 53;
Sbjct: 13 to 48


BLOCKS Protein Domain Analysis of NOV16a
AC#          Description                                          Strength  Score
BL00940B   0 Gamma-thionins family proteins.                        1324    1093
BL01102    0 Prokaryotic dksA/traR C4-type zinc finger.            1600    1053
BL00518    0 Zinc finger, C3HC4 type (RING finger), protei         1150    1034
BL01185D   0 C-terminal cystine knot proteins.                      1733    1026
BL00478A   0 LIM domain proteins.                                   1037    1023
BL00597B   0 Plant lipid transfer proteins.                        1514    1021
```

A PROSITE Protein Domain Matches analysis of the NOV16a protein suggests that NOV16a has one N-glycosylation site (Pattern-ID: ASN_ glycosylation PS00001 (Interpro)); six Protein kinase C phosphorylation sites (Pattern-ID: PKC_PHOSPHO_SITE PS00005 (Interpro)); three Casein kinase II phosphorylation sites (Pattern-ID: CK2_PHOSPHO_SITE PS00006 (Interpro)); three N-myristoylation sites (Pattern-ID: MYRISTYL PS00008 (Interpro)); and one Amidation site (Pattern-ID: AMIDATION PS00009 (Interpro)).

Table 16J lists the domain description from DOMAIN analysis results against NOV16b. This indicates that the NOV16b sequence has properties similar to those of other proteins known to contain this domain.

Table 16J. Domain Analysis of NOV16b

```
ProDom Analysis
prdm:3303 p36 (8) FGD1(2) - PROTEIN KINASE RHO/RAC FACTOR ZINC-FINGER PUTATIVE GUANINE
NUCLEOTIDE EXCHANGE GEF, 235 aa
Expect = 0.00014, identical = 20 of 50 (40%), positive = 24 of 50 (48%)

prdm:28902 p36 (1) YLN2_CAEEL - HYPOTHETICAL 46.2 KD TRP-ASP REPEATS CONTAINING
PROTEIN D2013.2 IN CHROMOSOME II. HYPOTHETICAL PROTEIN; REPEAT; WD REPEAT, 138 aa
Expect = 0.0018, identical = 14 of 38 (36%), positive = 18 of 38 (47%)

prdm:4778 p36 (5) - INHIBITOR SERINE PROTEASE CHYMOTRYPSIN/ELASTASE PROTEIN TRYPSIN
ISOINHIBITOR ISOINHIBITORS R10H1.1 CHROMOSOME, 67 aa
Expect = 0.051, identical = 14 of 36 (38%), positive = 21 of 36 (58%)

BLOCKS Protein Domain Analysis of NOV16b
AC#        Description                                        Strength  Score
BL00940B   Gamma-thionins family proteins.                      1324    1093
BL01102    Prokaryotic dksA/traR C4-type zinc finger.          1600    1053
BL00518    Zinc finger, C3HC4 type (RING finger), protei       1150    1034
BL01185D   C-terminal cystine knot proteins.                    1733    1026
BL00478A   LIM domain proteins                                 1037    1023
BL00597B   Plant lipid transfer proteins.                      1514    1021

PROSITE - Protein Domain Matches for Gene ID: NOV16-1
Pattern-ID: PKC_PHOSPHO_SITE PS00005 (Interpro) PDOC00005
6 Protein kinase C phosphorylation site

Pattern-ID: CK2_PHOSPHO_SITE PS00006 (Interpro) PDOC00006
3 Casein kinase II phosphorylation site

Pattern-ID: MYRISTYL PS00008 (Interpro) PDOC00008
2 N-myristoylation site
```

```
Pattern-ID: AMIDATION PS00009 (Interpro) PDOC00009
1 Amidation site

PFAM HMM Domain Analysis of NOV16b
Model            Domain  seq-f seq-t    hmm-f hmm-t       score  E-value
--------         ------- ----- -----    ----- -----       -----  -------
FYVE zinc finger 1/1       1    49 [.    13    66 ..     29.1   8.9e-07
```

In a BlastP analysis of a public database, NOV 16 a was found to have 70 of 70 (100%) identical to, and 70 of 70 (100%) positive with, the 146 aa Human ORFX ORF3149 polypeptide sequence SEQ ID NO:6298 (patp:AAB43385, Expect = 1.2e-36); 37 of 111 (33%) identical to, and 61 of 111 (54%) positive with, the 2052 aa Mus sp phosphatidylinositol-4-phosphate-5-kinase, designated p235 (patp:AAB08634, Expect = 6.9e-10); 21 of 47 (44%) identical to, and 25 of 47 (53%) positive with, the 195 aa Homo sapiens Polypeptide fragment encoded by gene 57 (patp:AAY01473, Expect = 3.5e-07); 28 of 64 (43%) identical to, and 37 of 64 (57%) positive with, the 1235 aa Xenopus sp Smad Anchor for Receptor Activation protein-1 (patp:AAY44751, Expect = 8.8e-07); and 18 of 47 (38%) identical to, and 24 of 47 (51%) positive with, the 138 aa Arabidopsis thaliana protein fragment SEQ ID NO: 28225 (patp:AAG24520, Expect = 3.3e-06). The Patp BLAST results for NOV16a and NOV16b are listed in Table 16K.

Table 16K. Patp alignments of NOV16

Sequences producing High-scoring Segment Pairs:	High Score	NOV16a Smallest Sum Prob. P(N)	NOV16b Smallest Sum Prob. (N)
patp:AAB43385 Human ORFX ORF3149 polypeptide sequence SEQ...	395	1.2e-36	1.2e-36
patp:AAB08634 A murine phosphatidylinositol-4-phosphate-5...	159	6.9e-10	6.9e-10
patp:AAY01473 Polypeptide fragment encoded by gene 57 - H...	129	3.5e-07	3.1e-07
patp:AAY44751 Xenopus Smad Anchor for Receptor Activation...	139	8.8e-07	6.7e-07
patp:AAG24520 Arabidopsis thaliana protein fragment SEQ I...	110	3.3e-06	3.0e-06
patp:AAY44749 Human Smad Anchor for Receptor Activation p...	134	4.6e-06	3.8e-06

The homologies shown above are shared by NOV16b insofar as NOV16b is homologous to NOV16a as shown in Table 16E.

Signaling by phosphorylated species of phosphatidylinositol (PI) appears to regulate diverse responses in eukaryotic cells. A differential display screen for fat- and muscle-specific transcripts led to identification and cloning of the full-length cDNA of a novel mammalian 2,052-amino-acid protein (p235) from a mouse adipocyte cDNA library. Analysis of the deduced amino acid sequence revealed that p235 contains an N-terminal zinc-binding FYVE finger, a chaperonin-like region in the middle of the molecule, and a consensus for phosphoinositide 5-kinases at the C terminus. p235 mRNA appears as a 9-kb transcript, enriched in insulin-sensitive cells and tissues, likely transcribed from a single-copy gene in at least two close-in-size splice

variants. Specific antibodies against mouse p235 were raised, and both the endogenously and heterologously expressed proteins were biochemically detected in 3T3-L1 adipocytes and transfected COS cells, respectively. Immunofluorescence microscopy analysis of endogenous p235 localization in 3T3-L1 adipocytes with affinity-purified anti-p235 antibodies documented a punctate peripheral pattern. In COS cells, the expressed p235 N-terminal but not the C-terminal region displayed a vesicular pattern similar to that in 3T3-L1 adipocytes that became diffuse upon Zn2+ chelation or FYVE finger truncation. A recombinant protein comprising the N-terminal but not the C-terminal region of the molecule was found to bind 2.2 mole equivalents of Zn2+. Determination of the lipid kinase activity in the p235 immunoprecipitates derived from 3T3-L1 adipocytes or from COS cells transiently expressing p235 revealed that p235 displayed unique preferences for PI substrate over already phosphorylated PI. In conclusion, the mouse p235 protein determines an important novel class of phosphoinositide kinases that seems to be targeted to specific intracellular loci by a Zn-dependent mechanism. See, PMID: 9858586

Isoforms of protein kinase B (PKB, or AKT1; 164730) are overexpressed in some ovarian, pancreatic, and breast cancer cells, and PKB has been shown to protect cells from apoptosis. Activation of PKB, which is preventable by inhibitors of phosphoinositide 3-kinase (see PIK3CG; 601232), is stimulated by insulin or growth factors after phosphorylation of PKB at thr308 and ser473. Alessi *et al.* (1997) biochemically purified a protein kinase, which they called PDK1, that phosphorylates PKB at thr308 in response to phosphotidylinositol 3,4,5-trisphosphate (PtdIns(3,4,5)P3) or phosphotidylinositol 3,4-biphosphate (PtdIns(3,4)P2) and enhances PKB activity. By microsequence analysis of the approximately 67- to 69-kD PDK1 protein, searching an EST database, and probing a breast cancer cell line cDNA library, Alessi *et al.* (1997) isolated a cDNA encoding PDK1, also called PDPK1. Sequence analysis predicted that the 556-amino acid PDPK1 protein contains a catalytic domain with 11 classic kinase subdomains and a C-terminal pleckstrin homology (PH) domain. Expression of recombinant PDPK1 resulted in the activation and phosphorylation of PKB at thr308 in a PtdIns(3,4,5)P3- or PtdIns(3,4)P2-dependent manner via the PH domains.

PtdIns(3,4,5)P3 and PtdIns(3,4)P2 bind to the PH domains of PKB and PDPK1, causing their translocation to the membrane and leading to PKB activation. See, Stephens *et al.*, Science 279: 710-714, 1998. PDPK1 selectively phosphorylates the 70-kD ribosomal protein S6 kinase (p70-RPS6K) at thr229, which is required for its activation. See, Pullen *et al.*, Science 279: 707-710, 1998. Thr229 of p70-RPS6K is homologous to thr308 of the PKB protein. The PDPK1 gene was mapped to 16p13.3 based on its identity to a sequence located in the same region as the PKD1 (601313) and TSC2 (191092) loci. See, Burn *et al.*, Genome Res. 6: 525-537, 1996; Alessi *et al.*, Curr. Biol. 7: 261-269, 1997; Alessi *et al.*, Curr. Biol. 7: 776-789, 1997.

The FYVE zinc finger is named after four proteins that it has been found in: Fab1, YOTB/ZK632.12, Vac1, and EEA1. The FYVE finger has been shown to bind two Zn2+ ions. The FYVE finger has eight potential zinc coordinating cysteine positions. Many members of this family also include two histidines in a motif R+HHC+XCG, where + represents a charged residue and X any residue. See, IPR000306

This indicates that the NOV16 sequence has properties similar to those of other proteins known to contain this/these domain(s) and similar to the properties of these domains.

The above defined information for this invention suggests that these FYVE finger-containing phosphoinositide kinase-like NOV16 proteins may function as a member of a "FYVE finger-containing phosphoinositide kinase-like protein family". Therefore, the NOV16 nucleic acids and proteins identified here may be useful in potential therapeutic applications implicated in (but not limited to) various pathologies and disorders as indicated below.

The protein similarity information, expression pattern, cellular localization, and map location for the protein and nucleic acid disclosed herein suggest that this FYVE finger-containing phosphoinositide kinase-like protein may have important structural and/or physiological functions characteristic of the FYVE finger-containing phosphoinositide kinase family. Therefore, the nucleic acids and proteins of the invention are useful in potential diagnostic and therapeutic applications and as a research tool. These include serving as a specific or selective nucleic acid or protein diagnostic and/or prognostic marker, wherein the presence or amount of the nucleic acid or the protein are to be assessed. These also include potential therapeutic applications such as the following: (i) a protein therapeutic, (ii) a small molecule drug target, (iii) an antibody target (therapeutic, diagnostic, drug targeting/cytotoxic antibody), (iv) a nucleic acid useful in gene therapy (gene delivery/gene ablation), (v) an agent promoting tissue regeneration *in vitro* and *in vivo*, and (vi) a biological defense weapon.

The nucleic acids and proteins of the invention have applications in the diagnosis and/or treatment of various diseases and disorders. For example, the compositions of the present invention will have efficacy for the treatment of patients suffering from: diabetes, obesity, fertility, signaling as well as other diseases, disorders and conditions.

Based on the tissues in which NOV16 is most highly expressed; including placenta, spleen, prostate, kidney, pancreas, thyroid, testis, ovary, uterus, heart, lung, brain cervix, umbilical vein, adrenal gland, bone and others; specific uses include developing products for the diagnosis or treatment of a variety of diseases and disorders. Additional disease indications and tissue expression for NOV16 is presented in Example 2.

These materials are further useful in the generation of antibodies that bind immunospecifically to the novel substances of the invention for use in diagnostic and/or

therapeutic methods. NOV16 nucleic acids and polypeptides are further useful in the generation of antibodies that bind immuno-specifically to the novel NOV16 substances for use in therapeutic or diagnostic methods. These antibodies may be generated according to methods known in the art, using prediction from hydrophobicity charts, as described in the "Anti-NOVX Antibodies" section below. For example the disclosed NOV16 proteins have multiple hydrophilic regions, each of which can be used as an immunogen. In one embodiment, a contemplated NOV16a epitope is from about amino acids 1 to 45. In additional embodiments, NOV16a epitopes are from about amino acids 50 to 60, from about amino acids 75 to 110, from about amino acids 120 to 160 and from about amino acids 190 to 206. In another embodiment, a NOV16b epitope is from about amino acids 1 to 45. In further embodiments, NOV16b epitopes are from about amino acids 50 to 70, from about amino acids 75 to 110, from about amino acids 120 to 160 and from about amino acids 180 to 202. This novel protein also has value in development of powerful assay system for functional analysis of various human disorders, which will help in understanding of pathology of the disease and development of new drug targets for various disorders.

NOVX Nucleic Acids and Polypeptides

One aspect of the invention pertains to isolated nucleic acid molecules that encode NOVX polypeptides or biologically active portions thereof. Also included in the invention are nucleic acid fragments sufficient for use as hybridization probes to identify NOVX-encoding nucleic acids (*e.g.*, NOVX mRNAs) and fragments for use as PCR primers for the amplification and/or mutation of NOVX nucleic acid molecules. As used herein, the term "nucleic acid molecule" is intended to include DNA molecules (*e.g.*, cDNA or genomic DNA), RNA molecules (*e.g.*, mRNA), analogs of the DNA or RNA generated using nucleotide analogs, and derivatives, fragments and homologs thereof. The nucleic acid molecule may be single-stranded or double-stranded, but preferably is comprised double-stranded DNA.

An NOVX nucleic acid can encode a mature NOVX polypeptide. As used herein, a "mature" form of a polypeptide or protein disclosed in the present invention is the product of a naturally occurring polypeptide or precursor form or proprotein. The naturally occurring polypeptide, precursor or proprotein includes, by way of nonlimiting example, the full-length gene product, encoded by the corresponding gene. Alternatively, it may be defined as the polypeptide, precursor or proprotein encoded by an ORF described herein. The product "mature" form arises, again by way of nonlimiting example, as a result of one or more naturally occurring processing steps as they may take place within the cell, or host cell, in which the gene product arises. Examples of such processing steps leading to a "mature" form of a polypeptide

or protein include the cleavage of the N-terminal methionine residue encoded by the initiation codon of an ORF, or the proteolytic cleavage of a signal peptide or leader sequence. Thus a mature form arising from a precursor polypeptide or protein that has residues 1 to N, where residue 1 is the N-terminal methionine, would have residues 2 through N remaining after removal of the N-terminal methionine. Alternatively, a mature form arising from a precursor polypeptide or protein having residues 1 to N, in which an N-terminal signal sequence from residue 1 to residue M is cleaved, would have the residues from residue M+1 to residue N remaining. Further as used herein, a "mature" form of a polypeptide or protein may arise from a step of post-translational modification other than a proteolytic cleavage event. Such additional processes include, by way of non-limiting example, glycosylation. myristoylation or phosphorylation. In general, a mature polypeptide or protein may result from the operation of only one of these processes, or a combination of any of them.

The term "probes", as utilized herein, refers to nucleic acid sequences of variable length, preferably between at least about 10 nucleotides (nt), 100 nt, or as many as approximately, *e.g.*, 6,000 nt, depending upon the specific use. Probes are used in the detection of identical, similar, or complementary nucleic acid sequences. Longer length probes are generally obtained from a natural or recombinant source, are highly specific, and much slower to hybridize than shorter-length oligomer probes. Probes may be single- or double-stranded and designed to have specificity in PCR, membrane-based hybridization technologies, or ELISA-like technologies.

The term "isolated" nucleic acid molecule, as utilized herein, is one, which is separated from other nucleic acid molecules which are present in the natural source of the nucleic acid. Preferably, an "isolated" nucleic acid is free of sequences which naturally flank the nucleic acid (*i.e.*, sequences located at the 5'- and 3'-termini of the nucleic acid) in the genomic DNA of the organism from which the nucleic acid is derived. For example, in various embodiments, the isolated NOVX nucleic acid molecules can contain less than about 5 kb, 4 kb, 3 kb, 2 kb, 1 kb, 0.5 kb or 0.1 kb of nucleotide sequences which naturally flank the nucleic acid molecule in genomic DNA of the cell/tissue from which the nucleic acid is derived (*e.g.*, brain, heart, liver, spleen, etc.). Moreover, an "isolated" nucleic acid molecule, such as a cDNA molecule, can be substantially free of other cellular material or culture medium when produced by recombinant techniques, or of chemical precursors or other chemicals when chemically synthesized.

A nucleic acid molecule of the invention, *e.g.*, a nucleic acid molecule having the nucleotide sequence SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102, or a complement of this aforementioned nucleotide sequence, can be isolated using standard molecular biology techniques and the sequence information provided herein. Using all or a portion of the nucleic acid sequence of SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51,

53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102 as a hybridization probe, NOVX molecules can be isolated using standard hybridization and cloning techniques (*e.g.*, as described in Sambrook, *et al.*, (eds.), MOLECULAR CLONING: A LABORATORY MANUAL 2nd Ed., Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY, 1989; and Ausubel, *et al.*, (eds.), CURRENT PROTOCOLS IN MOLECULAR BIOLOGY. John Wiley & Sons, New York, NY, 1993.)

A nucleic acid of the invention can be amplified using cDNA, mRNA or alternatively, genomic DNA, as a template and appropriate oligonucleotide primers according to standard PCR amplification techniques. The nucleic acid so amplified can be cloned into an appropriate vector and characterized by DNA sequence analysis. Furthermore, oligonucleotides corresponding to NOVX nucleotide sequences can be prepared by standard synthetic techniques, *e.g.*, using an automated DNA synthesizer.

As used herein, the term "oligonucleotide" refers to a series of linked nucleotide residues, which oligonucleotide has a sufficient number of nucleotide bases to be used in a PCR reaction. A short oligonucleotide sequence may be based on, or designed from, a genomic or cDNA sequence and is used to amplify, confirm, or reveal the presence of an identical, similar or complementary DNA or RNA in a particular cell or tissue. Oligonucleotides comprise portions of a nucleic acid sequence having about 10 nt, 50 nt, or 100 nt in length, preferably about 15 nt to 30 nt in length. In one embodiment of the invention, an oligonucleotide comprising a nucleic acid molecule less than 100 nt in length would further comprise at least 6 contiguous nucleotides SEQ ID NOS:2, 9, 11, 19, 27, 35, 43. 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102, or a complement thereof. Oligonucleotides may be chemically synthesized and may also be used as probes.

In another embodiment, an isolated nucleic acid molecule of the invention comprises a nucleic acid molecule that is a complement of the nucleotide sequence shown in SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102, or a portion of this nucleotide sequence (*e.g.*, a fragment that can be used as a probe or primer or a fragment encoding a biologically-active portion of an NOVX polypeptide). A nucleic acid molecule that is complementary to the nucleotide sequence shown SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 or 102is one that is sufficiently complementary to the nucleotide sequence shown SEQ ID NOS:2, 9, 11, 19. 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 or 102that it can hydrogen bond with little or no mismatches to the nucleotide sequence shown SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102, thereby forming a stable duplex.

As used herein, the term "complementary" refers to Watson-Crick or Hoogsteen base pairing between nucleotides units of a nucleic acid molecule, and the term "binding" means the

physical or chemical interaction between two polypeptides or compounds or associated polypeptides or compounds or combinations thereof. Binding includes ionic, non-ionic, van der Waals, hydrophobic interactions, and the like. A physical interaction can be either direct or indirect. Indirect interactions may be through or due to the effects of another polypeptide or compound. Direct binding refers to interactions that do not take place through, or due to, the effect of another polypeptide or compound, but instead are without other substantial chemical intermediates.

Fragments provided herein are defined as sequences of at least 6 (contiguous) nucleic acids or at least 4 (contiguous) amino acids, a length sufficient to allow for specific hybridization in the case of nucleic acids or for specific recognition of an epitope in the case of amino acids, respectively, and are at most some portion less than a full length sequence. Fragments may be derived from any contiguous portion of a nucleic acid or amino acid sequence of choice. Derivatives are nucleic acid sequences or amino acid sequences formed from the native compounds either directly or by modification or partial substitution. Analogs are nucleic acid sequences or amino acid sequences that have a structure similar to, but not identical to, the native compound but differs from it in respect to certain components or side chains. Analogs may be synthetic or from a different evolutionary origin and may have a similar or opposite metabolic activity compared to wild type. Homologs are nucleic acid sequences or amino acid sequences of a particular gene that are derived from different species.

Derivatives and analogs may be full length or other than full length, if the derivative or analog contains a modified nucleic acid or amino acid, as described below. Derivatives or analogs of the nucleic acids or proteins of the invention include, but are not limited to, molecules comprising regions that are substantially homologous to the nucleic acids or proteins of the invention, in various embodiments, by at least about 70%, 80%, or 95% identity (with a preferred identity of 80-95%) over a nucleic acid or amino acid sequence of identical size or when compared to an aligned sequence in which the alignment is done by a computer homology program known in the art, or whose encoding nucleic acid is capable of hybridizing to the complement of a sequence encoding the aforementioned proteins under stringent, moderately stringent, or low stringent conditions. *See e.g.* Ausubel. *et al.*, CURRENT PROTOCOLS IN MOLECULAR BIOLOGY, John Wiley & Sons, New York, NY, 1993, and below.

A "homologous nucleic acid sequence" or "homologous amino acid sequence," or variations thereof, refer to sequences characterized by a homology at the nucleotide level or amino acid level as discussed above. Homologous nucleotide sequences encode those sequences coding for isoforms of NOVX polypeptides. Isoforms can be expressed in different tissues of the same organism as a result of, for example, alternative splicing of RNA. Alternatively,

isoforms can be encoded by different genes. In the invention, homologous nucleotide sequences include nucleotide sequences encoding for an NOVX polypeptide of species other than humans, including, but not limited to: vertebrates, and thus can include, *e.g.*, frog, mouse, rat, rabbit, dog, cat cow, horse, and other organisms. Homologous nucleotide sequences also include, but are not limited to, naturally occurring allelic variations and mutations of the nucleotide sequences set forth herein. A homologous nucleotide sequence does not, however, include the exact nucleotide sequence encoding human NOVX protein. Homologous nucleic acid sequences include those nucleic acid sequences that encode conservative amino acid substitutions (see below) in SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102, as well as a polypeptide possessing NOVX biological activity. Various biological activities of the NOVX proteins are described below.

An NOVX polypeptide is encoded by the open reading frame ("ORF") of an NOVX nucleic acid. An ORF corresponds to a nucleotide sequence that could potentially be translated into a polypeptide. A stretch of nucleic acids comprising an ORF is uninterrupted by a stop codon. An ORF that represents the coding sequence for a full protein begins with an ATG "start" codon and terminates with one of the three "stop" codons, namely, TAA, TAG, or TGA. For the purposes of this invention, an ORF may be any part of a coding sequence, with or without a start codon, a stop codon, or both. For an ORF to be considered as a good candidate for coding for a *bona fide* cellular protein, a minimum size requirement is often set, *e.g.*, a stretch of DNA that would encode a protein of 50 amino acids or more.

The nucleotide sequences determined from the cloning of the human NOVX genes allows for the generation of probes and primers designed for use in identifying and/or cloning NOVX homologues in other cell types, *e.g.* from other tissues, as well as NOVX homologues from other vertebrates. The probe/primer typically comprises substantially purified oligonucleotide. The oligonucleotide typically comprises a region of nucleotide sequence that hybridizes under stringent conditions to at least about 12, 25, 50, 100, 150, 200, 250, 300, 350 or 400 consecutive sense strand nucleotide sequence SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 or 102; or an anti-sense strand nucleotide sequence of SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 or 102; or of a naturally occurring mutant of SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102.

Probes based on the human NOVX nucleotide sequences can be used to detect transcripts or genomic sequences encoding the same or homologous proteins. In various embodiments, the probe further comprises a label group attached thereto, *e.g.* the label group can be a radioisotope, a fluorescent compound, an enzyme, or an enzyme co-factor. Such probes can be used as a part

of a diagnostic test kit for identifying cells or tissues which mis-express an NOVX protein, such as by measuring a level of an NOVX-encoding nucleic acid in a sample of cells from a subject *e.g.*, detecting NOVX mRNA levels or determining whether a genomic NOVX gene has been mutated or deleted.

"A polypeptide having a biologically-active portion of an NOVX polypeptide" refers to polypeptides exhibiting activity similar, but not necessarily identical to, an activity of a polypeptide of the invention, including mature forms, as measured in a particular biological assay, with or without dose dependency. A nucleic acid fragment encoding a "biologically-active portion of NOVX" can be prepared by isolating a portion SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 or 102, that encodes a polypeptide having an NOVX biological activity (the biological activities of the NOVX proteins are described below), expressing the encoded portion of NOVX protein (*e.g.*, by recombinant expression *in vitro*) and assessing the activity of the encoded portion of NOVX.

NOVX Nucleic Acid and Polypeptide Variants

The invention further encompasses nucleic acid molecules that differ from the nucleotide sequences shown in SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102 due to degeneracy of the genetic code and thus encode the same NOVX proteins as that encoded by the nucleotide sequences shown in SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102. In another embodiment, an isolated nucleic acid molecule of the invention has a nucleotide sequence encoding a protein having an amino acid sequence shown in SEQ ID NOS:1, 8, 10, 12, 18, 20, 26, 28, 34, 36, 42, 44, 50, 52, 54, 60, 62, 64, 70, 72, 74, 76, 82, 89, 91, 99 or 101.

In addition to the human NOVX nucleotide sequences shown in SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102, it will be appreciated by those skilled in the art that DNA sequence polymorphisms that lead to changes in the amino acid sequences of the NOVX polypeptides may exist within a population (*e.g.*, the human population). Such genetic polymorphism in the NOVX genes may exist among individuals within a population due to natural allelic variation. As used herein, the terms "gene" and "recombinant gene" refer to nucleic acid molecules comprising an open reading frame (ORF) encoding an NOVX protein, preferably a vertebrate NOVX protein. Such natural allelic variations can typically result in 1-5% variance in the nucleotide sequence of the NOVX genes. Any and all such nucleotide variations and resulting amino acid polymorphisms in the NOVX polypeptides, which are the result of natural allelic variation and that do not alter the functional activity of the NOVX polypeptides, are intended to be within the scope of the invention.

Moreover, nucleic acid molecules encoding NOVX proteins from other species, and thus that have a nucleotide sequence that differs from the human SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102 are intended to be within the scope of the invention. Nucleic acid molecules corresponding to natural allelic variants and homologues of the NOVX cDNAs of the invention can be isolated based on their homology to the human NOVX nucleic acids disclosed herein using the human cDNAs, or a portion thereof, as a hybridization probe according to standard hybridization techniques under stringent hybridization conditions.

Accordingly, in another embodiment, an isolated nucleic acid molecule of the invention is at least 6 nucleotides in length and hybridizes under stringent conditions to the nucleic acid molecule comprising the nucleotide sequence of SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102. In another embodiment, the nucleic acid is at least 10, 25, 50, 100, 250, 500, 750. 1000, 1500. or 2000 or more nucleotides in length. In yet another embodiment, an isolated nucleic acid molecule of the invention hybridizes to the coding region. As used herein, the term "hybridizes under stringent conditions" is intended to describe conditions for hybridization and washing under which nucleotide sequences at least 60% homologous to each other typically remain hybridized to each other.

Homologs (*i.e.*, nucleic acids encoding NOVX proteins derived from species other than human) or other related sequences (*e.g.*, paralogs) can be obtained by low, moderate or high stringency hybridization with all or a portion of the particular human sequence as a probe using methods well known in the art for nucleic acid hybridization and cloning.

As used herein, the phrase "stringent hybridization conditions" refers to conditions under which a probe, primer or oligonucleotide will hybridize to its target sequence, but to no other sequences. Stringent conditions are sequence-dependent and will be different in different circumstances. Longer sequences hybridize specifically at higher temperatures than shorter sequences. Generally, stringent conditions are selected to be about 5 °C lower than the thermal melting point (Tm) for the specific sequence at a defined ionic strength and pH. The Tm is the temperature (under defined ionic strength, pH and nucleic acid concentration) at which 50% of the probes complementary to the target sequence hybridize to the target sequence at equilibrium. Since the target sequences are generally present at excess, at Tm, 50% of the probes are occupied at equilibrium. Typically, stringent conditions will be those in which the salt concentration is less than about 1.0 M sodium ion, typically about 0.01 to 1.0 M sodium ion (or other salts) at pH 7.0 to 8.3 and the temperature is at least about 30°C for short probes, primers or oligonucleotides (*e.g.*, 10 nt to 50 nt) and at least about 60°C for longer probes, primers and

oligonucleotides. Stringent conditions may also be achieved with the addition of destabilizing agents, such as formamide.

Stringent conditions are known to those skilled in the art and can be found in Ausubel, *et al.*, (eds.), CURRENT PROTOCOLS IN MOLECULAR BIOLOGY, John Wiley & Sons, N.Y. (1989), 6.3.1-6.3.6. Preferably, the conditions are such that sequences at least about 65%, 70%, 75%, 85%, 90%, 95%, 98%, or 99% homologous to each other typically remain hybridized to each other. A non-limiting example of stringent hybridization conditions are hybridization in a high salt buffer comprising 6X SSC, 50 mM Tris-HCl (pH 7.5), 1 mM EDTA, 0.02% PVP, 0.02% Ficoll, 0.02% BSA, and 500 mg/ml denatured salmon sperm DNA at 65°C, followed by one or more washes in 0.2X SSC, 0.01% BSA at 50°C. An isolated nucleic acid molecule of the invention that hybridizes under stringent conditions to the sequences SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102, corresponds to a naturally-occurring nucleic acid molecule. As used herein, a "naturally-occurring" nucleic acid molecule refers to an RNA or DNA molecule having a nucleotide sequence that occurs in nature (*e.g.*, encodes a natural protein).

In a second embodiment, a nucleic acid sequence that is hybridizable to the nucleic acid molecule comprising the nucleotide sequence of SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102, or fragments, analogs or derivatives thereof, under conditions of moderate stringency is provided. A non-limiting example of moderate stringency hybridization conditions are hybridization in 6X SSC, 5X Denhardt's solution, 0.5% SDS and 100 mg/ml denatured salmon sperm DNA at 55°C, followed by one or more washes in 1X SSC, 0.1% SDS at 37°C. Other conditions of moderate stringency that may be used are well-known within the art. *See, e.g.*, Ausubel, et *al.* (eds.), 1993, CURRENT PROTOCOLS IN MOLECULAR BIOLOGY, John Wiley & Sons, NY, and Kriegler, 1990; GENE TRANSFER AND EXPRESSION, A LABORATORY MANUAL, Stockton Press, NY.

In a third embodiment, a nucleic acid that is hybridizable to the nucleic acid molecule comprising the nucleotide sequences SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102, or fragments, analogs or derivatives thereof, under conditions of low stringency, is provided. A non-limiting example of low stringency hybridization conditions are hybridization in 35% formamide, 5X SSC, 50 mM Tris-HCl (pH 7.5), 5 mM EDTA, 0.02% PVP, 0.02% Ficoll, 0.2% BSA, 100 mg/ml denatured salmon sperm DNA, 10% (wt/vol) dextran sulfate at 40°C, followed by one or more washes in 2X SSC, 25 mM Tris-HCl (pH 7.4), 5 mM EDTA, and 0.1% SDS at 50°C. Other conditions of low stringency that may be used are well known in the art (*e.g.*, as employed for cross-species hybridizations). *See, e.g.,* Ausubel, *et al.* (eds.), 1993, CURRENT PROTOCOLS IN MOLECULAR BIOLOGY, John Wiley & Sons, NY, and

Kriegler, 1990, GENE TRANSFER AND EXPRESSION. A LABORATORY MANUAL, Stockton Press, NY; Shilo and Weinberg, 1981. *Proc Natl Acad Sci USA* 78: 6789-6792.

Conservative Mutations

In addition to naturally-occurring allelic variants of NOVX sequences that may exist in the population, the skilled artisan will further appreciate that changes can be introduced by mutation into the nucleotide sequences SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102, thereby leading to changes in the amino acid sequences of the encoded NOVX proteins, without altering the functional ability of said NOVX proteins. For example, nucleotide substitutions leading to amino acid substitutions at "non-essential" amino acid residues can be made in the sequence SEQ ID NOS:1, 8, 10, 12, 18, 20, 26, 28, 34, 36, 42, 44, 50, 52, 54, 60, 62, 64, 70, 72, 74, 76, 82, 89, 91, 99 or 101. A "non-essential" amino acid residue is a residue that can be altered from the wild-type sequences of the NOVX proteins without altering their biological activity, whereas an "essential" amino acid residue is required for such biological activity. For example, amino acid residues that are conserved among the NOVX proteins of the invention are predicted to be particularly non-amenable to alteration. Amino acids for which conservative substitutions can be made are well-known within the art.

Another aspect of the invention pertains to nucleic acid molecules encoding NOVX proteins that contain changes in amino acid residues that are not essential for activity. Such NOVX proteins differ in amino acid sequence from SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102 yet retain biological activity. In one embodiment, the isolated nucleic acid molecule comprises a nucleotide sequence encoding a protein, wherein the protein comprises an amino acid sequence at least about 45% homologous to the amino acid sequences SEQ ID NOS:1, 8, 10, 12, 18, 20, 26, 28, 34, 36, 42, 44, 50, 52, 54, 60, 62, 64, 70, 72, 74, 76, 82, 89, 91, 99 and 101. Preferably, the protein encoded by the nucleic acid molecule is at least about 60% homologous to SEQ ID NOS:1, 8, 10, 12, 18, 20, 26, 28, 34, 36, 42, 44, 50, 52, 54, 60, 62, 64, 70, 72, 74, 76, 82, 89, 91, 99 and 101; more preferably at least about 70% homologous SEQ ID NOS:1, 8, 10, 12, 18, 20, 26, 28, 34, 36, 42, 44, 50, 52, 54, 60, 62, 64, 70, 72, 74, 76, 82, 89, 91, 99 or 101; still more preferably at least about 80% homologous to SEQ ID NOS:1, 8, 10, 12, 18, 20, 26, 28, 34, 36, 42, 44, 50, 52, 54, 60, 62, 64, 70, 72, 74, 76, 82, 89, 91, 99 or 101; even more preferably at least about 90% homologous to SEQ ID NOS:1, 8, 10, 12, 18, 20, 26, 28, 34, 36, 42, 44, 50, 52, 54, 60, 62, 64, 70, 72, 74, 76, 82, 89, 91, 99 or 101; and most preferably at least about 95% homologous to SEQ ID NOS:1, 8, 10, 12, 18, 20, 26, 28, 34, 36, 42, 44, 50, 52, 54, 60, 62, 64, 70, 72, 74, 76, 82, 89, 91, 99 or 101.

An isolated nucleic acid molecule encoding an NOVX protein homologous to the protein of SEQ ID NOS:1, 8, 10, 12, 18, 20, 26, 28, 34, 36, 42, 44, 50, 52, 54, 60, 62, 64, 70, 72, 74, 76, 82, 89, 91, 99 or 101 can be created by introducing one or more nucleotide substitutions, additions or deletions into the nucleotide sequence of SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102, such that one or more amino acid substitutions, additions or deletions are introduced into the encoded protein.

Mutations can be introduced into SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102 by standard techniques, such as site-directed mutagenesis and PCR-mediated mutagenesis. Preferably, conservative amino acid substitutions are made at one or more predicted, non-essential amino acid residues. A "conservative amino acid substitution" is one in which the amino acid residue is replaced with an amino acid residue having a similar side chain. Families of amino acid residues having similar side chains have been defined within the art. These families include amino acids with basic side chains (*e.g.*, lysine, arginine, histidine), acidic side chains (*e.g.*, aspartic acid, glutamic acid), uncharged polar side chains (*e.g.*, glycine, asparagine, glutamine, serine, threonine, tyrosine, cysteine), nonpolar side chains (*e.g.*, alanine, valine, leucine, isoleucine, proline, phenylalanine, methionine, tryptophan), beta-branched side chains (*e.g.*, threonine, valine, isoleucine) and aromatic side chains (*e.g.*, tyrosine, phenylalanine, tryptophan, histidine). Thus, a predicted non-essential amino acid residue in the NOVX protein is replaced with another amino acid residue from the same side chain family. Alternatively, in another embodiment, mutations can be introduced randomly along all or part of an NOVX coding sequence, such as by saturation mutagenesis, and the resultant mutants can be screened for NOVX biological activity to identify mutants that retain activity. Following mutagenesis SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102, the encoded protein can be expressed by any recombinant technology known in the art and the activity of the protein can be determined.

The relatedness of amino acid families may also be determined based on side chain interactions. Substituted amino acids may be fully conserved "strong" residues or fully conserved "weak" residues. The "strong" group of conserved amino acid residues may be any one of the following groups: STA, NEQK, NHQK, NDEQ, QHRK, MILV, MILF, HY, FYW, wherein the single letter amino acid codes are grouped by those amino acids that may be substituted for each other. Likewise, the "weak" group of conserved residues may be any one of the following: CSA, ATV, SAG, STNK, STPA, SGND, SNDEQK, NDEQHK, NEQHRK, VLIM, HFY, wherein the letters within each group represent the single letter amino acid code.

In one embodiment, a mutant NOVX protein can be assayed for (*i*) the ability to form protein:protein interactions with other NOVX proteins, other cell-surface proteins, or

biologically-active portions thereof. (*ii*) complex formation between a mutant NOVX protein and an NOVX ligand; or (*iii*) the ability of a mutant NOVX protein to bind to an intracellular target protein or biologically-active portion thereof; (*e.g.* avidin proteins).

In yet another embodiment, a mutant NOVX protein can be assayed for the ability to regulate a specific biological function (*e.g.*, regulation of insulin release).

Antisense Nucleic Acids

Another aspect of the invention pertains to isolated antisense nucleic acid molecules that are hybridizable to or complementary to the nucleic acid molecule comprising the nucleotide sequence of SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102, or fragments, analogs or derivatives thereof. An "antisense" nucleic acid comprises a nucleotide sequence that is complementary to a "sense" nucleic acid encoding a protein (*e.g.*, complementary to the coding strand of a double-stranded cDNA molecule or complementary to an mRNA sequence). In specific aspects, antisense nucleic acid molecules are provided that comprise a sequence complementary to at least about 10, 25, 50, 100, 250 or 500 nucleotides or an entire NOVX coding strand, or to only a portion thereof. Nucleic acid molecules encoding fragments, homologs, derivatives and analogs of an NOVX protein of SEQ ID NOS:1, 8, 10, 12, 18, 20, 26, 28, 34, 36, 42, 44, 50, 52, 54, 60, 62, 64, 70, 72, 74, 76, 82, 89, 91, 99 or 101, or antisense nucleic acids complementary to an NOVX nucleic acid sequence of SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102, are additionally provided.

In one embodiment, an antisense nucleic acid molecule is antisense to a "coding region" of the coding strand of a nucleotide sequence encoding an NOVX protein. The term "coding region" refers to the region of the nucleotide sequence comprising codons which are translated into amino acid residues. In another embodiment, the antisense nucleic acid molecule is antisense to a "noncoding region" of the coding strand of a nucleotide sequence encoding the NOVX protein. The term "noncoding region" refers to 5' and 3' sequences which flank the coding region that are not translated into amino acids (*i.e.*, also referred to as 5' and 3' untranslated regions).

Given the coding strand sequences encoding the NOVX protein disclosed herein, antisense nucleic acids of the invention can be designed according to the rules of Watson and Crick or Hoogsteen base pairing. The antisense nucleic acid molecule can be complementary to the entire coding region of NOVX mRNA, but more preferably is an oligonucleotide that is antisense to only a portion of the coding or noncoding region of NOVX mRNA. For example, the antisense oligonucleotide can be complementary to the region surrounding the translation

start site of NOVX mRNA. An antisense oligonucleotide can be, for example, about 5, 10, 15, 20, 25, 30, 35, 40, 45 or 50 nucleotides in length. An antisense nucleic acid of the invention can be constructed using chemical synthesis or enzymatic ligation reactions using procedures known in the art. For example, an antisense nucleic acid (*e.g.*, an antisense oligonucleotide) can be chemically synthesized using naturally-occurring nucleotides or variously modified nucleotides designed to increase the biological stability of the molecules or to increase the physical stability of the duplex formed between the antisense and sense nucleic acids (*e.g.*, phosphorothioate derivatives and acridine substituted nucleotides can be used).

Examples of modified nucleotides that can be used to generate the antisense nucleic acid include: 5-fluorouracil, 5-bromouracil, 5-chlorouracil, 5-iodouracil, hypoxanthine, xanthine, 4-acetylcytosine, 5-(carboxyhydroxylmethyl) uracil, 5-carboxymethylaminomethyl-2-thiouridine, 5-carboxymethylaminomethyluracil, dihydrouracil, beta-D-galactosylqueosine, inosine, N6-isopentenyladenine, 1-methylguanine, 1-methylinosine, 2,2-dimethylguanine, 2-methyladenine, 2-methylguanine, 3-methylcytosine, 5-methylcytosine, N6-adenine, 7-methylguanine, 5-methylaminomethyluracil, 5-methoxyaminomethyl-2-thiouracil, beta-D-mannosylqueosine, 5'-methoxycarboxymethyluracil, 5-methoxyuracil, 2-methylthio-N6-isopentenyladenine, uracil-5-oxyacetic acid (v), wybutoxosine, pseudouracil, queosine, 2-thiocytosine, 5-methyl-2-thiouracil, 2-thiouracil, 4-thiouracil, 5-methyluracil, uracil-5-oxyacetic acid methylester, uracil-5-oxyacetic acid (v), 5-methyl-2-thiouracil, 3-(3-amino-3-N-2-carboxypropyl) uracil, (acp3)w, and 2,6-diaminopurine. Alternatively, the antisense nucleic acid can be produced biologically using an expression vector into which a nucleic acid has been subcloned in an antisense orientation (*i.e.*, RNA transcribed from the inserted nucleic acid will be of an antisense orientation to a target nucleic acid of interest, described further in the following subsection).

The antisense nucleic acid molecules of the invention are typically administered to a subject or generated *in situ* such that they hybridize with or bind to cellular mRNA and/or genomic DNA encoding an NOVX protein to thereby inhibit expression of the protein (*e.g.*, by inhibiting transcription and/or translation). The hybridization can be by conventional nucleotide complementarity to form a stable duplex, or, for example, in the case of an antisense nucleic acid molecule that binds to DNA duplexes, through specific interactions in the major groove of the double helix. An example of a route of administration of antisense nucleic acid molecules of the invention includes direct injection at a tissue site. Alternatively, antisense nucleic acid molecules can be modified to target selected cells and then administered systemically. For example, for systemic administration, antisense molecules can be modified such that they specifically bind to receptors or antigens expressed on a selected cell surface (*e.g.*, by linking the

antisense nucleic acid molecules to peptides or antibodies that bind to cell surface receptors or antigens). The antisense nucleic acid molecules can also be delivered to cells using the vectors described herein. To achieve sufficient nucleic acid molecules, vector constructs in which the antisense nucleic acid molecule is placed under the control of a strong pol II or pol III promoter are preferred.

In yet another embodiment, the antisense nucleic acid molecule of the invention is an α-anomeric nucleic acid molecule. An α-anomeric nucleic acid molecule forms specific double-stranded hybrids with complementary RNA in which, contrary to the usual β-units, the strands run parallel to each other. *See, e.g.*, Gaultier, *et al.*, 1987. *Nucl. Acids Res.* 15: 6625-6641. The antisense nucleic acid molecule can also comprise a 2'-o-methylribonucleotide (*See, e.g.*, Inoue, *et al.* 1987. *Nucl. Acids Res.* 15: 6131-6148) or a chimeric RNA-DNA analogue (*See, e.g.*, Inoue, *et al.*, 1987. *FEBS Lett.* 215: 327-330.

Ribozymes and PNA Moieties

Nucleic acid modifications include, by way of non-limiting example, modified bases, and nucleic acids whose sugar phosphate backbones are modified or derivatized. These modifications are carried out at least in part to enhance the chemical stability of the modified nucleic acid, such that they may be used, for example, as antisense binding nucleic acids in therapeutic applications in a subject.

In one embodiment, an antisense nucleic acid of the invention is a ribozyme. Ribozymes are catalytic RNA molecules with ribonuclease activity that are capable of cleaving a single-stranded nucleic acid, such as an mRNA, to which they have a complementary region. Thus, ribozymes (*e.g.*, hammerhead ribozymes as described in Haselhoff and Gerlach 1988. *Nature* 334: 585-591) can be used to catalytically cleave NOVX mRNA transcripts to thereby inhibit translation of NOVX mRNA. A ribozyme having specificity for an NOVX-encoding nucleic acid can be designed based upon the nucleotide sequence of an NOVX cDNA disclosed herein (*i.e.*, SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102). For example, a derivative of a *Tetrahymena* L-19 IVS RNA can be constructed in which the nucleotide sequence of the active site is complementary to the nucleotide sequence to be cleaved in an NOVX-encoding mRNA. *See, e g.*, U.S. Patent 4,987,071 to Cech, *et al.* and U.S. Patent 5,116,742 to Cech, *et al.* NOVX mRNA can also be used to select a catalytic RNA having a specific ribonuclease activity from a pool of RNA molecules. *See, e.g.*, Bartel *et al.*, (1993) *Science* 261:1411-1418.

Alternatively, NOVX gene expression can be inhibited by targeting nucleotide sequences complementary to the regulatory region of the NOVX nucleic acid (*e.g.*, the NOVX promoter

and/or enhancers) to form triple helical structures that prevent transcription of the NOVX gene in target cells. *See, e.g.,* Helene, 1991. *Anticancer Drug Des.* 6: 569-84; Helene, *et al.* 1992. *Ann. N.Y. Acad. Sci.* 660: 27-36; Maher, 1992. *Bioassays* 14: 807-15.

In various embodiments, the NOVX nucleic acids can be modified at the base moiety, sugar moiety or phosphate backbone to improve, *e.g.*, the stability, hybridization, or solubility of the molecule. For example, the deoxyribose phosphate backbone of the nucleic acids can be modified to generate peptide nucleic acids. *See, e.g.,* Hyrup, *et al.*, 1996. *Bioorg Med Chem* 4: 5-23. As used herein, the terms "peptide nucleic acids" or "PNAs" refer to nucleic acid mimics (*e.g.*, DNA mimics) in which the deoxyribose phosphate backbone is replaced by a pseudopeptide backbone and only the four natural nucleobases are retained. The neutral backbone of PNAs has been shown to allow for specific hybridization to DNA and RNA under conditions of low ionic strength. The synthesis of PNA oligomers can be performed using standard solid phase peptide synthesis protocols as described in Hyrup, *et al.*, 1996. *supra*; Perry-O'Keefe, *et al.*, 1996. *Proc. Natl. Acad. Sci. USA* 93: 14670-14675.

PNAs of NOVX can be used in therapeutic and diagnostic applications. For example, PNAs can be used as antisense or antigene agents for sequence-specific modulation of gene expression by, *e.g.*, inducing transcription or translation arrest or inhibiting replication. PNAs of NOVX can also be used, for example, in the analysis of single base pair mutations in a gene (*e.g.*, PNA directed PCR clamping; as artificial restriction enzymes when used in combination with other enzymes, *e.g.*, S_1 nucleases (*See,* Hyrup, *et al.,* 1996.*supra*); or as probes or primers for DNA sequence and hybridization (*See,* Hyrup, *et al.*, 1996, *supra*; Perry-O'Keefe, *et al.,* 1996. *supra*).

In another embodiment, PNAs of NOVX can be modified, *e.g.*, to enhance their stability or cellular uptake, by attaching lipophilic or other helper groups to PNA, by the formation of PNA-DNA chimeras, or by the use of liposomes or other techniques of drug delivery known in the art. For example, PNA-DNA chimeras of NOVX can be generated that may combine the advantageous properties of PNA and DNA. Such chimeras allow DNA recognition enzymes (*e.g.*, RNase H and DNA polymerases) to interact with the DNA portion while the PNA portion would provide high binding affinity and specificity. PNA-DNA chimeras can be linked using linkers of appropriate lengths selected in terms of base stacking, number of bonds between the nucleobases, and orientation (*see,* Hyrup, *et al.*, 1996. *supra*). The synthesis of PNA-DNA chimeras can be performed as described in Hyrup, *et al.,* 1996. *supra* and Finn, *et al.*, 1996. *Nucl Acids Res* 24: 3357-3363. For example, a DNA chain can be synthesized on a solid support using standard phosphoramidite coupling chemistry, and modified nucleoside analogs, *e.g.*, 5'-(4-methoxytrityl)amino-5'-deoxy-thymidine phosphoramidite, can be used between the PNA

and the 5' end of DNA. *See, e.g.,* Mag, *et al.*, 1989. *Nucl Acid Res* 17: 5973-5988. PNA monomers are then coupled in a stepwise manner to produce a chimeric molecule with a 5' PNA segment and a 3' DNA segment. *See, e.g.,* Finn, *et al.*, 1996. *supra.* Alternatively, chimeric molecules can be synthesized with a 5' DNA segment and a 3' PNA segment. *See, e.g.,* Petersen, *et al.*, 1975. *Bioorg. Med Chem. Lett* 5: 1119-11124.

In other embodiments, the oligonucleotide may include other appended groups such as peptides (*e.g.*, for targeting host cell receptors *in vivo*), or agents facilitating transport across the cell membrane (*see, e.g.*, Letsinger, *et al.*, 1989. *Proc. Natl. Acad. Sci. U.S.A.* 86: 6553-6556; Lemaitre, *et al.*, 1987. *Proc. Natl. Acad. Sci.* 84: 648-652; PCT Publication No. WO88/09810) or the blood-brain barrier (*see, e.g.*, PCT Publication No. WO 89/10134). In addition, oligonucleotides can be modified with hybridization triggered cleavage agents (*see, e.g.,* Krol, *et al.*, 1988. *BioTechniques* 6:958-976) or intercalating agents (*see, e.g.*, Zon, 1988. *Pharm. Res.* 5: 539-549). To this end, the oligonucleotide may be conjugated to another molecule, *e.g.*, a peptide, a hybridization triggered cross-linking agent, a transport agent, a hybridization-triggered cleavage agent, and the like.

NOVX Polypeptides

A polypeptide according to the invention includes a polypeptide including the amino acid sequence of NOVX polypeptides whose sequences are provided in SEQ ID NOS:1, 8, 10, 12, 18, 20, 26, 28, 34, 36, 42, 44, 50, 52, 54, 60, 62, 64, 70, 72, 74, 76, 82, 89, 91, 99 or 101. The invention also includes a mutant or variant protein any of whose residues may be changed from the corresponding residues shown in SEQ ID NOS:1, 8, 10, 12, 18, 20, 26, 28, 34, 36, 42, 44, 50, 52, 54, 60, 62, 64, 70, 72, 74, 76, 82, 89, 91, 99 or 101 while still encoding a protein that maintains its NOVX activities and physiological functions, or a functional fragment thereof.

In general, an NOVX variant that preserves NOVX-like function includes any variant in which residues at a particular position in the sequence have been substituted by other amino acids, and further include the possibility of inserting an additional residue or residues between two residues of the parent protein as well as the possibility of deleting one or more residues from the parent sequence. Any amino acid substitution, insertion, or deletion is encompassed by the invention. In favorable circumstances, the substitution is a conservative substitution as defined above.

One aspect of the invention pertains to isolated NOVX proteins, and biologically-active portions thereof, or derivatives, fragments, analogs or homologs thereof. Also provided are polypeptide fragments suitable for use as immunogens to raise anti-NOVX antibodies. In one embodiment, native NOVX proteins can be isolated from cells or tissue sources by an

appropriate purification scheme using standard protein purification techniques. In another embodiment, NOVX proteins are produced by recombinant DNA techniques. Alternative to recombinant expression, an NOVX protein or polypeptide can be synthesized chemically using standard peptide synthesis techniques.

An "isolated" or "purified" polypeptide or protein or biologically-active portion thereof is substantially free of cellular material or other contaminating proteins from the cell or tissue source from which the NOVX protein is derived, or substantially free from chemical precursors or other chemicals when chemically synthesized. The language "substantially free of cellular material" includes preparations of NOVX proteins in which the protein is separated from cellular components of the cells from which it is isolated or recombinantly-produced. In one embodiment, the language "substantially free of cellular material" includes preparations of NOVX proteins having less than about 30% (by dry weight) of non-NOVX proteins (also referred to herein as a "contaminating protein"), more preferably less than about 20% of non-NOVX proteins, still more preferably less than about 10% of non-NOVX proteins, and most preferably less than about 5% of non-NOVX proteins. When the NOVX protein or biologically-active portion thereof is recombinantly-produced, it is also preferably substantially free of culture medium, *i.e.*, culture medium represents less than about 20%, more preferably less than about 10%, and most preferably less than about 5% of the volume of the NOVX protein preparation.

The language "substantially free of chemical precursors or other chemicals" includes preparations of NOVX proteins in which the protein is separated from chemical precursors or other chemicals that are involved in the synthesis of the protein. In one embodiment, the language "substantially free of chemical precursors or other chemicals" includes preparations of NOVX proteins having less than about 30% (by dry weight) of chemical precursors or non-NOVX chemicals, more preferably less than about 20% chemical precursors or non-NOVX chemicals, still more preferably less than about 10% chemical precursors or non-NOVX chemicals, and most preferably less than about 5% chemical precursors or non-NOVX chemicals.

Biologically-active portions of NOVX proteins include peptides comprising amino acid sequences sufficiently homologous to or derived from the amino acid sequences of the NOVX proteins (*e.g.*, the amino acid sequence shown in SEQ ID NOS:1, 8, 10, 12, 18, 20, 26, 28, 34, 36, 42, 44, 50, 52, 54, 60, 62, 64, 70, 72, 74, 76, 82, 89, 91, 99 or 101) that include fewer amino acids than the full-length NOVX proteins, and exhibit at least one activity of an NOVX protein. Typically, biologically-active portions comprise a domain or motif with at least one activity of

the NOVX protein. A biologically-active portion of an NOVX protein can be a polypeptide which is, for example, 10, 25, 50, 100 or more amino acid residues in length.

Moreover, other biologically-active portions, in which other regions of the protein are deleted, can be prepared by recombinant techniques and evaluated for one or more of the functional activities of a native NOVX protein.

In an embodiment, the NOVX protein has an amino acid sequence shown SEQ ID NOS:1, 8, 10, 12, 18, 20, 26, 28, 34, 36, 42, 44, 50, 52, 54, 60, 62, 64, 70, 72, 74, 76, 82, 89, 91, 99 or 101. In other embodiments, the NOVX protein is substantially homologous to SEQ ID NOS:1, 8, 10, 12, 18, 20, 26, 28, 34, 36, 42, 44, 50, 52, 54, 60, 62, 64, 70, 72, 74, 76, 82, 89, 91, 99 or 101, and retains the functional activity of the protein of SEQ ID NOS:1, 8, 10, 12, 18, 20, 26, 28, 34, 36, 42, 44, 50, 52, 54, 60, 62, 64, 70, 72, 74, 76, 82, 89, 91, 99 or 101, yet differs in amino acid sequence due to natural allelic variation or mutagenesis, as described in detail, below. Accordingly, in another embodiment, the NOVX protein is a protein that comprises an amino acid sequence at least about 45% homologous to the amino acid sequence SEQ ID NOS:1, 8, 10, 12, 18, 20, 26, 28, 34, 36, 42, 44, 50, 52, 54, 60, 62, 64, 70, 72, 74, 76, 82, 89, 91, 99 or 101, and retains the functional activity of the NOVX proteins of SEQ ID NOS:1, 8, 10, 12, 18, 20, 26, 28, 34, 36, 42, 44, 50, 52, 54, 60, 62, 64, 70, 72, 74, 76, 82, 89, 91, 99 or 101.

Determining Homology Between Two or More Sequences

To determine the percent homology of two amino acid sequences or of two nucleic acids, the sequences are aligned for optimal comparison purposes (*e.g.*, gaps can be introduced in the sequence of a first amino acid or nucleic acid sequence for optimal alignment with a second amino or nucleic acid sequence). The amino acid residues or nucleotides at corresponding amino acid positions or nucleotide positions are then compared. When a position in the first sequence is occupied by the same amino acid residue or nucleotide as the corresponding position in the second sequence, then the molecules are homologous at that position (*i.e.*, as used herein amino acid or nucleic acid "homology" is equivalent to amino acid or nucleic acid "identity").

The nucleic acid sequence homology may be determined as the degree of identity between two sequences. The homology may be determined using computer programs known in the art, such as GAP software provided in the GCG program package. *See,* Needleman and Wunsch, 1970. *J Mol Biol* 48: 443-453. Using GCG GAP software with the following settings for nucleic acid sequence comparison: GAP creation penalty of 5.0 and GAP extension penalty of 0.3, the coding region of the analogous nucleic acid sequences referred to above exhibits a degree of identity preferably of at least 70%, 75%, 80%, 85%, 90%, 95%, 98%, or 99%, with the

CDS (encoding) part of the DNA sequence shown in SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102.

The term "sequence identity" refers to the degree to which two polynucleotide or polypeptide sequences are identical on a residue-by-residue basis over a particular region of comparison. The term "percentage of sequence identity" is calculated by comparing two optimally aligned sequences over that region of comparison, determining the number of positions at which the identical nucleic acid base (*e.g.*, A, T, C, G, U, or I, in the case of nucleic acids) occurs in both sequences to yield the number of matched positions, dividing the number of matched positions by the total number of positions in the region of comparison (*i.e.*, the window size), and multiplying the result by 100 to yield the percentage of sequence identity. The term "substantial identity" as used herein denotes a characteristic of a polynucleotide sequence, wherein the polynucleotide comprises a sequence that has at least 80 percent sequence identity, preferably at least 85 percent identity and often 90 to 95 percent sequence identity, more usually at least 99 percent sequence identity as compared to a reference sequence over a comparison region.

Chimeric and Fusion Proteins

The invention also provides NOVX chimeric or fusion proteins. As used herein, an NOVX "chimeric protein" or "fusion protein" comprises an NOVX polypeptide operatively-linked to a non-NOVX polypeptide. An "NOVX polypeptide" refers to a polypeptide having an amino acid sequence corresponding to an NOVX protein SEQ ID NOS:1, 8, 10, 12, 18, 20, 26, 28, 34, 36, 42, 44, 50, 52, 54, 60, 62, 64, 70, 72, 74, 76, 82, 89, 91, 99 or 101), whereas a "non-NOVX polypeptide" refers to a polypeptide having an amino acid sequence corresponding to a protein that is not substantially homologous to the NOVX protein, *e.g.*, a protein that is different from the NOVX protein and that is derived from the same or a different organism. Within an NOVX fusion protein the NOVX polypeptide can correspond to all or a portion of an NOVX protein. In one embodiment, an NOVX fusion protein comprises at least one biologically-active portion of an NOVX protein. In another embodiment, an NOVX fusion protein comprises at least two biologically-active portions of an NOVX protein. In yet another embodiment, an NOVX fusion protein comprises at least three biologically-active portions of an NOVX protein. Within the fusion protein, the term "operatively-linked" is intended to indicate that the NOVX polypeptide and the non-NOVX polypeptide are fused in-frame with one another. The non-NOVX polypeptide can be fused to the N-terminus or C-terminus of the NOVX polypeptide.

In one embodiment, the fusion protein is a GST-NOVX fusion protein in which the NOVX sequences are fused to the C-terminus of the GST (glutathione S-transferase) sequences. Such fusion proteins can facilitate the purification of recombinant NOVX polypeptides.

In another embodiment, the fusion protein is an NOVX protein containing a heterologous signal sequence at its N-terminus. In certain host cells (*e.g.*, mammalian host cells), expression and/or secretion of NOVX can be increased through use of a heterologous signal sequence.

In yet another embodiment, the fusion protein is an NOVX-immunoglobulin fusion protein in which the NOVX sequences are fused to sequences derived from a member of the immunoglobulin protein family. The NOVX-immunoglobulin fusion proteins of the invention can be incorporated into pharmaceutical compositions and administered to a subject to inhibit an interaction between an NOVX ligand and an NOVX protein on the surface of a cell, to thereby suppress NOVX-mediated signal transduction *in vivo*. The NOVX-immunoglobulin fusion proteins can be used to affect the bioavailability of an NOVX cognate ligand. Inhibition of the NOVX ligand/NOVX interaction may be useful therapeutically for both the treatment of proliferative and differentiative disorders, as well as modulating (*e.g.* promoting or inhibiting) cell survival. Moreover, the NOVX-immunoglobulin fusion proteins of the invention can be used as immunogens to produce anti-NOVX antibodies in a subject, to purify NOVX ligands, and in screening assays to identify molecules that inhibit the interaction of NOVX with an NOVX ligand.

An NOVX chimeric or fusion protein of the invention can be produced by standard recombinant DNA techniques. For example, DNA fragments coding for the different polypeptide sequences are ligated together in-frame in accordance with conventional techniques, *e.g.*, by employing blunt-ended or stagger-ended termini for ligation, restriction enzyme digestion to provide for appropriate termini, filling-in of cohesive ends as appropriate, alkaline phosphatase treatment to avoid undesirable joining, and enzymatic ligation. In another embodiment, the fusion gene can be synthesized by conventional techniques including automated DNA synthesizers. Alternatively, PCR amplification of gene fragments can be carried out using anchor primers that give rise to complementary overhangs between two consecutive gene fragments that can subsequently be annealed and reamplified to generate a chimeric gene sequence (*see, e.g.,* Ausubel, *et al.* (eds.) CURRENT PROTOCOLS IN MOLECULAR BIOLOGY, John Wiley & Sons, 1992). Moreover, many expression vectors are commercially available that already encode a fusion moiety (*e.g.*, a GST polypeptide). An NOVX-encoding nucleic acid can be cloned into such an expression vector such that the fusion moiety is linked in-frame to the NOVX protein.

NOVX Agonists and Antagonists

The invention also pertains to variants of the NOVX proteins that function as either NOVX agonists (*i.e.*, mimetics) or as NOVX antagonists. Variants of the NOVX protein can be generated by mutagenesis (*e.g.*, discrete point mutation or truncation of the NOVX protein). An agonist of the NOVX protein can retain substantially the same, or a subset of, the biological activities of the naturally occurring form of the NOVX protein. An antagonist of the NOVX protein can inhibit one or more of the activities of the naturally occurring form of the NOVX protein by, for example, competitively binding to a downstream or upstream member of a cellular signaling cascade which includes the NOVX protein. Thus, specific biological effects can be elicited by treatment with a variant of limited function. In one embodiment, treatment of a subject with a variant having a subset of the biological activities of the naturally occurring form of the protein has fewer side effects in a subject relative to treatment with the naturally occurring form of the NOVX proteins.

Variants of the NOVX proteins that function as either NOVX agonists (*i.e.*, mimetics) or as NOVX antagonists can be identified by screening combinatorial libraries of mutants (*e.g.*, truncation mutants) of the NOVX proteins for NOVX protein agonist or antagonist activity. In one embodiment, a variegated library of NOVX variants is generated by combinatorial mutagenesis at the nucleic acid level and is encoded by a variegated gene library. A variegated library of NOVX variants can be produced by, for example, enzymatically ligating a mixture of synthetic oligonucleotides into gene sequences such that a degenerate set of potential NOVX sequences is expressible as individual polypeptides, or alternatively, as a set of larger fusion proteins (*e.g.*, for phage display) containing the set of NOVX sequences therein. There are a variety of methods which can be used to produce libraries of potential NOVX variants from a degenerate oligonucleotide sequence. Chemical synthesis of a degenerate gene sequence can be performed in an automatic DNA synthesizer, and the synthetic gene then ligated into an appropriate expression vector. Use of a degenerate set of genes allows for the provision, in one mixture, of all of the sequences encoding the desired set of potential NOVX sequences. Methods for synthesizing degenerate oligonucleotides are well-known within the art. *See, e.g.*, Narang, 1983. *Tetrahedron* 39: 3; Itakura, *et al.*, 1984. *Annu. Rev. Biochem.* 53: 323; Itakura, *et al.*, 1984. *Science* 198: 1056; Ike, *et al.*, 1983. *Nucl. Acids Res.* 11: 477.

Polypeptide Libraries

In addition, libraries of fragments of the NOVX protein coding sequences can be used to generate a variegated population of NOVX fragments for screening and subsequent selection of

variants of an NOVX protein. In one embodiment, a library of coding sequence fragments can be generated by treating a double stranded PCR fragment of an NOVX coding sequence with a nuclease under conditions wherein nicking occurs only about once per molecule, denaturing the double stranded DNA, renaturing the DNA to form double-stranded DNA that can include sense/antisense pairs from different nicked products, removing single stranded portions from reformed duplexes by treatment with S_1 nuclease, and ligating the resulting fragment library into an expression vector. By this method, expression libraries can be derived which encodes N-terminal and internal fragments of various sizes of the NOVX proteins.

Various techniques are known in the art for screening gene products of combinatorial libraries made by point mutations or truncation, and for screening cDNA libraries for gene products having a selected property. Such techniques are adaptable for rapid screening of the gene libraries generated by the combinatorial mutagenesis of NOVX proteins. The most widely used techniques, which are amenable to high throughput analysis, for screening large gene libraries typically include cloning the gene library into replicable expression vectors, transforming appropriate cells with the resulting library of vectors, and expressing the combinatorial genes under conditions in which detection of a desired activity facilitates isolation of the vector encoding the gene whose product was detected. Recursive ensemble mutagenesis (REM), a new technique that enhances the frequency of functional mutants in the libraries, can be used in combination with the screening assays to identify NOVX variants. *See, e.g.,* Arkin and Yourvan, 1992. *Proc. Natl. Acad. Sci. USA* 89: 7811-7815; Delgrave, *et al.*, 1993. *Protein Engineering* 6:327-331.

Anti-NOVX Antibodies

Also included in the invention are antibodies to NOVX proteins, or fragments of NOVX proteins. The term "antibody" as used herein refers to immunoglobulin molecules and immunologically active portions of immunoglobulin (Ig) molecules, *i.e.*, molecules that contain an antigen binding site that specifically binds (immunoreacts with) an antigen. Such antibodies include, but are not limited to, polyclonal, monoclonal, chimeric, single chain, F_{ab}, $F_{ab'}$ and $F_{(ab')2}$ fragments, and an F_{ab} expression library. In general, an antibody molecule obtained from humans relates to any of the classes IgG, IgM, IgA, IgE and IgD, which differ from one another by the nature of the heavy chain present in the molecule. Certain classes have subclasses as well, such as IgG_1, IgG_2, and others. Furthermore, in humans, the light chain may be a kappa chain or a lambda chain. Reference herein to antibodies includes a reference to all such classes, subclasses and types of human antibody species.

An isolated NOVX-related protein of the invention may be intended to serve as an antigen, or a portion or fragment thereof, and additionally can be used as an immunogen to generate antibodies that immunospecifically bind the antigen, using standard techniques for polyclonal and monoclonal antibody preparation. The full-length protein can be used or, alternatively, the invention provides antigenic peptide fragments of the antigen for use as immunogens. An antigenic peptide fragment comprises at least 6 amino acid residues of the amino acid sequence of the full length protein and encompasses an epitope thereof such that an antibody raised against the peptide forms a specific immune complex with the full length protein or with any fragment that contains the epitope. Preferably, the antigenic peptide comprises at least 10 amino acid residues, or at least 15 amino acid residues, or at least 20 amino acid residues, or at least 30 amino acid residues. Preferred epitopes encompassed by the antigenic peptide are regions of the protein that are located on its surface; commonly these are hydrophilic regions.

In certain embodiments of the invention, at least one epitope encompassed by the antigenic peptide is a region of NOVX-related protein that is located on the surface of the protein, *e.g.*, a hydrophilic region. A hydrophobicity analysis of the human NOVX-related protein sequence will indicate which regions of a NOVX-related protein are particularly hydrophilic and, therefore, are likely to encode surface residues useful for targeting antibody production. As a means for targeting antibody production, hydropathy plots showing regions of hydrophilicity and hydrophobicity may be generated by any method well known in the art, including, for example, the Kyte Doolittle or the Hopp Woods methods, either with or without Fourier transformation. See, *e.g.*, Hopp and Woods, 1981, *Proc. Nat. Acad. Sci. USA* 78: 3824-3828; Kyte and Doolittle 1982, *J. Mol. Biol.* 157: 105-142, each of which is incorporated herein by reference in its entirety. Antibodies that are specific for one or more domains within an antigenic protein, or derivatives, fragments, analogs or homologs thereof, are also provided herein.

A protein of the invention, or a derivative, fragment, analog, homolog or ortholog thereof, may be utilized as an immunogen in the generation of antibodies that immunospecifically bind these protein components.

Various procedures known within the art may be used for the production of polyclonal or monoclonal antibodies directed against a protein of the invention, or against derivatives, fragments, analogs homologs or orthologs thereof (see, for example, Antibodies: A Laboratory Manual, Harlow and Lane, 1988, Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY, incorporated herein by reference). Some of these antibodies are discussed below.

Polyclonal Antibodies

For the production of polyclonal antibodies, various suitable host animals (*e.g.*, rabbit, goat, mouse or other mammal) may be immunized by one or more injections with the native protein, a synthetic variant thereof, or a derivative of the foregoing. An appropriate immunogenic preparation can contain, for example, the naturally occurring immunogenic protein, a chemically synthesized polypeptide representing the immunogenic protein, or a recombinantly expressed immunogenic protein. Furthermore, the protein may be conjugated to a second protein known to be immunogenic in the mammal being immunized. Examples of such immunogenic proteins include but are not limited to keyhole limpet hemocyanin, serum albumin, bovine thyroglobulin, and soybean trypsin inhibitor. The preparation can further include an adjuvant. Various adjuvants used to increase the immunological response include, but are not limited to, Freund's (complete and incomplete), mineral gels (*e.g.*, aluminum hydroxide), surface active substances (*e.g.*, lysolecithin, pluronic polyols, polyanions, peptides, oil emulsions, dinitrophenol, etc.), adjuvants usable in humans such as Bacille Calmette-Guerin and Corynebacterium parvum, or similar immunostimulatory agents. Additional examples of adjuvants which can be employed include MPL-TDM adjuvant (monophosphoryl Lipid A, synthetic trehalose dicorynomycolate).

The polyclonal antibody molecules directed against the immunogenic protein can be isolated from the mammal (*e.g.*, from the blood) and further purified by well known techniques, such as affinity chromatography using protein A or protein G, which provide primarily the IgG fraction of immune serum. Subsequently, or alternatively, the specific antigen which is the target of the immunoglobulin sought, or an epitope thereof, may be immobilized on a column to purify the immune specific antibody by immunoaffinity chromatography. Purification of immunoglobulins is discussed, for example, by D. Wilkinson (The Scientist, published by The Scientist, Inc., Philadelphia PA, Vol. 14, No. 8 (April 17, 2000), pp. 25-28).

Monoclonal Antibodies

The term "monoclonal antibody" (MAb) or "monoclonal antibody composition", as used herein, refers to a population of antibody molecules that contain only one molecular species of antibody molecule consisting of a unique light chain gene product and a unique heavy chain gene product. In particular, the complementarity determining regions (CDRs) of the monoclonal antibody are identical in all the molecules of the population. MAbs thus contain an antigen binding site capable of immunoreacting with a particular epitope of the antigen characterized by a unique binding affinity for it.

Monoclonal antibodies can be prepared using hybridoma methods, such as those described by Kohler and Milstein, *Nature*, 256:495 (1975). In a hybridoma method, a mouse, hamster, or other appropriate host animal, is typically immunized with an immunizing agent to

elicit lymphocytes that produce or are capable of producing antibodies that will specifically bind to the immunizing agent. Alternatively, the lymphocytes can be immunized in vitro.

The immunizing agent will typically include the protein antigen, a fragment thereof or a fusion protein thereof. Generally, either peripheral blood lymphocytes are used if cells of human origin are desired, or spleen cells or lymph node cells are used if non-human mammalian sources are desired. The lymphocytes are then fused with an immortalized cell line using a suitable fusing agent, such as polyethylene glycol, to form a hybridoma cell (Goding, MONOCLONAL ANTIBODIES: PRINCIPLES AND PRACTICE, Academic Press, (1986) pp. 59-103). Immortalized cell lines are usually transformed mammalian cells, particularly myeloma cells of rodent, bovine and human origin. Usually, rat or mouse myeloma cell lines are employed. The hybridoma cells can be cultured in a suitable culture medium that preferably contains one or more substances that inhibit the growth or survival of the unfused, immortalized cells. For example, if the parental cells lack the enzyme hypoxanthine guanine phosphoribosyl transferase (HGPRT or HPRT), the culture medium for the hybridomas typically will include hypoxanthine, aminopterin, and thymidine ("HAT medium"), which substances prevent the growth of HGPRT-deficient cells.

Preferred immortalized cell lines are those that fuse efficiently, support stable high level expression of antibody by the selected antibody-producing cells, and are sensitive to a medium such as HAT medium. More preferred immortalized cell lines are murine myeloma lines, which can be obtained, for instance, from the Salk Institute Cell Distribution Center, San Diego, California and the American Type Culture Collection, Manassas, Virginia. Human myeloma and mouse-human heteromyeloma cell lines also have been described for the production of human monoclonal antibodies (Kozbor, *J. Immunol.*, 133:3001 (1984); Brodeur *et al.*, MONOCLONAL ANTIBODY PRODUCTION TECHNIQUES AND APPLICATIONS, Marcel Dekker, Inc., New York, (1987) pp. 51-63).

The culture medium in which the hybridoma cells are cultured can then be assayed for the presence of monoclonal antibodies directed against the antigen. Preferably, the binding specificity of monoclonal antibodies produced by the hybridoma cells is determined by immunoprecipitation or by an in vitro binding assay, such as radioimmunoassay (RIA) or enzyme-linked immunoabsorbent assay (ELISA). Such techniques and assays are known in the art. The binding affinity of the monoclonal antibody can, for example, be determined by the Scatchard analysis of Munson and Pollard, *Anal. Biochem.*, 107:220 (1980). Preferably, antibodies having a high degree of specificity and a high binding affinity for the target antigen are isolated.

After the desired hybridoma cells are identified, the clones can be subcloned by limiting dilution procedures and grown by standard methods. Suitable culture media for this purpose

include, for example, Dulbecco's Modified Eagle's Medium and RPMI-1640 medium. Alternatively, the hybridoma cells can be grown in vivo as ascites in a mammal.

The monoclonal antibodies secreted by the subclones can be isolated or purified from the culture medium or ascites fluid by conventional immunoglobulin purification procedures such as, for example, protein A-Sepharose, hydroxylapatite chromatography, gel electrophoresis, dialysis, or affinity chromatography.

The monoclonal antibodies can also be made by recombinant DNA methods, such as those described in U.S. Patent No. 4,816,567. DNA encoding the monoclonal antibodies of the invention can be readily isolated and sequenced using conventional procedures (*e.g.*, by using oligonucleotide probes that are capable of binding specifically to genes encoding the heavy and light chains of murine antibodies). The hybridoma cells of the invention serve as a preferred source of such DNA. Once isolated, the DNA can be placed into expression vectors, which are then transfected into host cells such as simian COS cells, Chinese hamster ovary (CHO) cells, or myeloma cells that do not otherwise produce immunoglobulin protein, to obtain the synthesis of monoclonal antibodies in the recombinant host cells. The DNA also can be modified, for example, by substituting the coding sequence for human heavy and light chain constant domains in place of the homologous murine sequences (U.S. Patent No. 4,816,567; Morrison, *Nature* 368, 812-13 (1994)) or by covalently joining to the immunoglobulin coding sequence all or part of the coding sequence for a non-immunoglobulin polypeptide. Such a non-immunoglobulin polypeptide can be substituted for the constant domains of an antibody of the invention, or can be substituted for the variable domains of one antigen-combining site of an antibody of the invention to create a chimeric bivalent antibody.

Humanized Antibodies

The antibodies directed against the protein antigens of the invention can further comprise humanized antibodies or human antibodies. These antibodies are suitable for administration to humans without engendering an immune response by the human against the administered immunoglobulin. Humanized forms of antibodies are chimeric immunoglobulins, immunoglobulin chains or fragments thereof (such as Fv, Fab, Fab', $F(ab')_2$ or other antigen-binding subsequences of antibodies) that are principally comprised of the sequence of a human immunoglobulin, and contain minimal sequence derived from a non-human immunoglobulin. Humanization can be performed following the method of Winter and co-workers (Jones *et al.*, *Nature*, 321:522-525 (1986); Riechmann *et al.*, *Nature*, 332:323-327 (1988); Verhoeyen *et al.*, *Science*, 239:1534-1536 (1988)), by substituting rodent CDRs or CDR sequences for the corresponding sequences of a human antibody. (See also U.S. Patent No. 5,225,539.) In some instances, Fv framework residues of the human immunoglobulin are replaced by corresponding

non-human residues. Humanized antibodies can also comprise residues which are found neither in the recipient antibody nor in the imported CDR or framework sequences. In general, the humanized antibody will comprise substantially all of at least one, and typically two, variable domains, in which all or substantially all of the CDR regions correspond to those of a non-human immunoglobulin and all or substantially all of the framework regions are those of a human immunoglobulin consensus sequence. The humanized antibody optimally also will comprise at least a portion of an immunoglobulin constant region (Fc), typically that of a human immunoglobulin (Jones *et al.*, 1986; Riechmann *et al.*, 1988; and Presta, *Curr. Op. Struct. Biol.*, 2:593-596 (1992)).

Human Antibodies

Fully human antibodies relate to antibody molecules in which essentially the entire sequences of both the light chain and the heavy chain, including the CDRs, arise from human genes. Such antibodies are termed "human antibodies", or "fully human antibodies" herein. Human monoclonal antibodies can be prepared by the trioma technique; the human B-cell hybridoma technique (see Kozbor, *et al.*, 1983 Immunol Today 4: 72) and the EBV hybridoma technique to produce human monoclonal antibodies (see Cole, *et al.*, 1985 In: MONOCLONAL ANTIBODIES AND CANCER THERAPY, Alan R. Liss, Inc., pp. 77-96). Human monoclonal antibodies may be utilized in the practice of the present invention and may be produced by using human hybridomas (see Cote, *et al.*, 1983. Proc Natl Acad Sci USA 80: 2026-2030) or by transforming human B-cells with Epstein Barr Virus in vitro (see Cole, *et al.*, 1985 In: MONOCLONAL ANTIBODIES AND CANCER THERAPY, Alan R. Liss, Inc., pp. 77-96).

In addition, human antibodies can also be produced using additional techniques, including phage display libraries (Hoogenboom and Winter, *J. Mol. Biol.*, 227:381 (1991); Marks *et al.*, *J. Mol. Biol.*, 222:581 (1991)). Similarly, human antibodies can be made by introducing human immunoglobulin loci into transgenic animals, *e.g.*, mice in which the endogenous immunoglobulin genes have been partially or completely inactivated. Upon challenge, human antibody production is observed, which closely resembles that seen in humans in all respects, including gene rearrangement, assembly, and antibody repertoire. This approach is described, for example, in U.S. Patent Nos. 5,545,807; 5,545,806; 5,569,825; 5,625,126; 5,633,425; 5,661,016, and in Marks *et al.* (*Bio/Technology* 10, 779-783 (1992)); Lonberg *et al.* (*Nature* 368 856-859 (1994)); Morrison (*Nature* 368, 812-13 (1994)); Fishwild *et al*,(*Nature Biotechnology* 14, 845-51 (1996)); Neuberger (*Nature Biotechnology* 14, 826 (1996)); and Lonberg and Huszar (*Intern. Rev. Immunol.* 13 65-93 (1995)).

Human antibodies may additionally be produced using transgenic nonhuman animals which are modified so as to produce fully human antibodies rather than the animal's endogenous

antibodies in response to challenge by an antigen. (See PCT publication WO94/02602). The endogenous genes encoding the heavy and light immunoglobulin chains in the nonhuman host have been incapacitated, and active loci encoding human heavy and light chain immunoglobulins are inserted into the host's genome. The human genes are incorporated, for example, using yeast artificial chromosomes containing the requisite human DNA segments. An animal which provides all the desired modifications is then obtained as progeny by crossbreeding intermediate transgenic animals containing fewer than the full complement of the modifications. The preferred embodiment of such a nonhuman animal is a mouse, and is termed the Xenomouse™ as disclosed in PCT publications WO 96/33735 and WO 96/34096. This animal produces B cells which secrete fully human immunoglobulins. The antibodies can be obtained directly from the animal after immunization with an immunogen of interest, as, for example, a preparation of a polyclonal antibody, or alternatively from immortalized B cells derived from the animal, such as hybridomas producing monoclonal antibodies. Additionally, the genes encoding the immunoglobulins with human variable regions can be recovered and expressed to obtain the antibodies directly, or can be further modified to obtain analogs of antibodies such as, for example, single chain Fv molecules.

An example of a method of producing a nonhuman host, exemplified as a mouse, lacking expression of an endogenous immunoglobulin heavy chain is disclosed in U.S. Patent No. 5,939,598. It can be obtained by a method including deleting the J segment genes from at least one endogenous heavy chain locus in an embryonic stem cell to prevent rearrangement of the locus and to prevent formation of a transcript of a rearranged immunoglobulin heavy chain locus, the deletion being effected by a targeting vector containing a gene encoding a selectable marker; and producing from the embryonic stem cell a transgenic mouse whose somatic and germ cells contain the gene encoding the selectable marker.

A method for producing an antibody of interest, such as a human antibody, is disclosed in U.S. Patent No. 5,916,771. It includes introducing an expression vector that contains a nucleotide sequence encoding a heavy chain into one mammalian host cell in culture, introducing an expression vector containing a nucleotide sequence encoding a light chain into another mammalian host cell, and fusing the two cells to form a hybrid cell. The hybrid cell expresses an antibody containing the heavy chain and the light chain.

In a further improvement on this procedure, a method for identifying a clinically relevant epitope on an immunogen, and a correlative method for selecting an antibody that binds immunospecifically to the relevant epitope with high affinity, are disclosed in PCT publication WO 99/53049.

F_{ab} Fragments and Single Chain Antibodies

According to the invention, techniques can be adapted for the production of single-chain antibodies specific to an antigenic protein of the invention (see *e.g.*, U.S. Patent No. 4,946,778). In addition, methods can be adapted for the construction of F_{ab} expression libraries (see *e.g.*, Huse, *et al.*, 1989 Science 246: 1275-1281) to allow rapid and effective identification of monoclonal F_{ab} fragments with the desired specificity for a protein or derivatives, fragments, analogs or homologs thereof. Antibody fragments that contain the idiotypes to a protein antigen may be produced by techniques known in the art including, but not limited to: (i) an $F_{(ab')2}$ fragment produced by pepsin digestion of an antibody molecule; (ii) an F_{ab} fragment generated by reducing the disulfide bridges of an $F_{(ab')2}$ fragment; (iii) an F_{ab} fragment generated by the treatment of the antibody molecule with papain and a reducing agent and (iv) F_v fragments.

Bispecific Antibodies

Bispecific antibodies are monoclonal, preferably human or humanized, antibodies that have binding specificities for at least two different antigens. In the present case, one of the binding specificities is for an antigenic protein of the invention. The second binding target is any other antigen, and advantageously is a cell-surface protein or receptor or receptor subunit.

Methods for making bispecific antibodies are known in the art. Traditionally, the recombinant production of bispecific antibodies is based on the co-expression of two immunoglobulin heavy-chain/light-chain pairs, where the two heavy chains have different specificities (Milstein and Cuello, *Nature*, 305:537-539 (1983)). Because of the random assortment of immunoglobulin heavy and light chains, these hybridomas (quadromas) produce a potential mixture of ten different antibody molecules, of which only one has the correct bispecific structure. The purification of the correct molecule is usually accomplished by affinity chromatography steps. Similar procedures are disclosed in WO 93/08829, published 13 May 1993, and in Traunecker *et al.*, 1991 *EMBO J.*, 10:3655-3659.

Antibody variable domains with the desired binding specificities (antibody-antigen combining sites) can be fused to immunoglobulin constant domain sequences. The fusion preferably is with an immunoglobulin heavy-chain constant domain, comprising at least part of the hinge, CH2, and CH3 regions. It is preferred to have the first heavy-chain constant region (CH1) containing the site necessary for light-chain binding present in at least one of the fusions. DNAs encoding the immunoglobulin heavy-chain fusions and, if desired, the immunoglobulin light chain, are inserted into separate expression vectors, and are co-transfected into a suitable host organism. For further details of generating bispecific antibodies see, for example, Suresh *et al.*, *Methods in Enzymology*, 121:210 (1986).

According to another approach described in WO 96/27011, the interface between a pair of antibody molecules can be engineered to maximize the percentage of heterodimers which are

recovered from recombinant cell culture. The preferred interface comprises at least a part of the CH3 region of an antibody constant domain. In this method, one or more small amino acid side chains from the interface of the first antibody molecule are replaced with larger side chains (*e.g.* tyrosine or tryptophan). Compensatory "cavities" of identical or similar size to the large side chain(s) are created on the interface of the second antibody molecule by replacing large amino acid side chains with smaller ones (*e.g.* alanine or threonine). This provides a mechanism for increasing the yield of the heterodimer over other unwanted end-products such as homodimers.

Bispecific antibodies can be prepared as full length antibodies or antibody fragments (*e.g.* $F(ab')_2$ bispecific antibodies). Techniques for generating bispecific antibodies from antibody fragments have been described in the literature. For example, bispecific antibodies can be prepared using chemical linkage. Brennan *et al.*, *Science* 229:81 (1985) describe a procedure wherein intact antibodies are proteolytically cleaved to generate $F(ab')_2$ fragments. These fragments are reduced in the presence of the dithiol complexing agent sodium arsenite to stabilize vicinal dithiols and prevent intermolecular disulfide formation. The Fab' fragments generated are then converted to thionitrobenzoate (TNB) derivatives. One of the Fab'-TNB derivatives is then reconverted to the Fab'-thiol by reduction with mercaptoethylamine and is mixed with an equimolar amount of the other Fab'-TNB derivative to form the bispecific antibody. The bispecific antibodies produced can be used as agents for the selective immobilization of enzymes.

Additionally, Fab' fragments can be directly recovered from E. coli and chemically coupled to form bispecific antibodies. Shalaby *et al.*, *J Exp. Med.* 175:217-225 (1992) describe the production of a fully humanized bispecific antibody $F(ab')_2$ molecule. Each Fab' fragment was separately secreted from E. coli and subjected to directed chemical coupling in vitro to form the bispecific antibody. The bispecific antibody thus formed was able to bind to cells overexpressing the ErbB2 receptor and normal human T cells, as well as trigger the lytic activity of human cytotoxic lymphocytes against human breast tumor targets.

Various techniques for making and isolating bispecific antibody fragments directly from recombinant cell culture have also been described. For example, bispecific antibodies have been produced using leucine zippers. Kostelny *et al.*, *J. Immunol.* 148(5):1547-1553 (1992). The leucine zipper peptides from the Fos and Jun proteins were linked to the Fab' portions of two different antibodies by gene fusion. The antibody homodimers were reduced at the hinge region to form monomers and then re-oxidized to form the antibody heterodimers. This method can also be utilized for the production of antibody homodimers. The "diabody" technology described by Hollinger *et al.*, *Proc. Natl. Acad. Sci. USA* 90:6444-6448 (1993) has provided an alternative mechanism for making bispecific antibody fragments. The fragments comprise a

heavy-chain variable domain (V_H) connected to a light-chain variable domain (V_L) by a linker which is too short to allow pairing between the two domains on the same chain. Accordingly, the V_H and V_L domains of one fragment are forced to pair with the complementary V_L and V_H domains of another fragment, thereby forming two antigen-binding sites. Another strategy for making bispecific antibody fragments by the use of single-chain Fv (sFv) dimers has also been reported. See, Gruber *et al.*, *J. Immunol.* 152:5368 (1994).

Antibodies with more than two valencies are contemplated. For example, trispecific antibodies can be prepared. Tutt *et al.*, *J. Immunol.* 147:60 (1991).

Exemplary bispecific antibodies can bind to two different epitopes, at least one of which originates in the protein antigen of the invention. Alternatively, an anti-antigenic arm of an immunoglobulin molecule can be combined with an arm which binds to a triggering molecule on a leukocyte such as a T-cell receptor molecule (*e.g.* CD2, CD3, CD28, or B7), or Fc receptors for IgG (FcγR), such as FcγRI (CD64), FcγRII (CD32) and FcγRIII (CD16) so as to focus cellular defense mechanisms to the cell expressing the particular antigen. Bispecific antibodies can also be used to direct cytotoxic agents to cells which express a particular antigen. These antibodies possess an antigen-binding arm and an arm which binds a cytotoxic agent or a radionuclide chelator, such as EOTUBE, DPTA, DOTA, or TETA. Another bispecific antibody of interest binds the protein antigen described herein and further binds tissue factor (TF).

Heteroconjugate Antibodies

Heteroconjugate antibodies are also within the scope of the present invention. Heteroconjugate antibodies are composed of two covalently joined antibodies. Such antibodies have, for example, been proposed to target immune system cells to unwanted cells (U.S. Patent No. 4,676,980), and for treatment of HIV infection (WO 91/00360; WO 92/200373; EP 03089). It is contemplated that the antibodies can be prepared in vitro using known methods in synthetic protein chemistry, including those involving crosslinking agents. For example, immunotoxins can be constructed using a disulfide exchange reaction or by forming a thioether bond. Examples of suitable reagents for this purpose include iminothiolate and methyl-4-mercaptobutyrimidate and those disclosed, for example, in U.S. Patent No. 4,676,980.

Effector Function Engineering

It can be desirable to modify the antibody of the invention with respect to effector function, so as to enhance, *e.g.*, the effectiveness of the antibody in treating cancer. For example, cysteine residue(s) can be introduced into the Fc region, thereby allowing interchain disulfide bond formation in this region. The homodimeric antibody thus generated can have improved internalization capability and/or increased complement-mediated cell killing and antibody-dependent cellular cytotoxicity (ADCC). See Caron *et al.*, J. Exp Med., 176: 1191-

1195 (1992) and Shopes, J. Immunol., 148: 2918-2922 (1992). Homodimeric antibodies with enhanced anti-tumor activity can also be prepared using heterobifunctional cross-linkers as described in Wolff *et al.* Cancer Research, 53: 2560-2565 (1993). Alternatively, an antibody can be engineered that has dual Fc regions and can thereby have enhanced complement lysis and ADCC capabilities. See Stevenson *et al.*, Anti-Cancer Drug Design, 3: 219-230 (1989).

Immunoconjugates

The invention also pertains to immunoconjugates comprising an antibody conjugated to a cytotoxic agent such as a chemotherapeutic agent, toxin (*e.g.*, an enzymatically active toxin of bacterial, fungal, plant, or animal origin, or fragments thereof), or a radioactive isotope (*i.e.*, a radioconjugate).

Chemotherapeutic agents useful in the generation of such immunoconjugates have been described above. Enzymatically active toxins and fragments thereof that can be used include diphtheria A chain, nonbinding active fragments of diphtheria toxin, exotoxin A chain (from Pseudomonas aeruginosa), ricin A chain, abrin A chain, modeccin A chain, alpha-sarcin, Aleurites fordii proteins, dianthin proteins, Phytolaca americana proteins (PAPI, PAPII, and PAP-S), momordica charantia inhibitor, curcin, crotin, sapaonaria officinalis inhibitor, gelonin, mitogellin, restrictocin, phenomycin, enomycin, and the tricothecenes. A variety of radionuclides are available for the production of radioconjugated antibodies. Examples include ^{212}Bi, ^{131}I, ^{131}In, ^{90}Y, and ^{186}Re.

Conjugates of the antibody and cytotoxic agent are made using a variety of bifunctional protein-coupling agents such as N-succinimidyl-3-(2-pyridyldithiol) propionate (SPDP), iminothiolane (IT), bifunctional derivatives of imidoesters (such as dimethyl adipimidate HCL), active esters (such as disuccinimidyl suberate), aldehydes (such as glutareldehyde), bis-azido compounds (such as bis (p-azidobenzoyl) hexanediamine), bis-diazonium derivatives (such as bis-(p-diazoniumbenzoyl)-ethylenediamine), diisocyanates (such as tolyene 2,6-diisocyanate), and bis-active fluorine compounds (such as 1,5-difluoro-2,4-dinitrobenzene). For example, a ricin immunotoxin can be prepared as described in Vitetta *et al.*, Science, 238: 1098 (1987). Carbon-14-labeled 1-isothiocyanatobenzyl-3-methyldiethylene triaminepentaacetic acid (MX-DTPA) is an exemplary chelating agent for conjugation of radionucleotide to the antibody. See WO94/11026.

In another embodiment, the antibody can be conjugated to a "receptor" (such streptavidin) for utilization in tumor pretargeting wherein the antibody-receptor conjugate is administered to the patient, followed by removal of unbound conjugate from the circulation using a clearing agent and then administration of a "ligand" (*e.g.*, avidin) that is in turn conjugated to a cytotoxic agent.

In one embodiment, methods for the screening of antibodies that possess the desired specificity include, but are not limited to, enzyme-linked immunosorbent assay (ELISA) and other immunologically-mediated techniques known within the art. In a specific embodiment, selection of antibodies that are specific to a particular domain of an NOVX protein is facilitated by generation of hybridomas that bind to the fragment of an NOVX protein possessing such a domain. Thus, antibodies that are specific for a desired domain within an NOVX protein, or derivatives, fragments, analogs or homologs thereof, are also provided herein.

Anti-NOVX antibodies may be used in methods known within the art relating to the localization and/or quantitation of an NOVX protein (*e.g.*, for use in measuring levels of the NOVX protein within appropriate physiological samples, for use in diagnostic methods, for use in imaging the protein, and the like). In a given embodiment, antibodies for NOVX proteins, or derivatives, fragments, analogs or homologs thereof, that contain the antibody derived binding domain, are utilized as pharmacologically-active compounds (hereinafter "Therapeutics").

An anti-NOVX antibody (*e.g.*, monoclonal antibody) can be used to isolate an NOVX polypeptide by standard techniques, such as affinity chromatography or immunoprecipitation. An anti-NOVX antibody can facilitate the purification of natural NOVX polypeptide from cells and of recombinantly-produced NOVX polypeptide expressed in host cells. Moreover, an anti-NOVX antibody can be used to detect NOVX protein (*e.g.*, in a cellular lysate or cell supernatant) in order to evaluate the abundance and pattern of expression of the NOVX protein. Anti-NOVX antibodies can be used diagnostically to monitor protein levels in tissue as part of a clinical testing procedure, *e.g.*, to, for example, determine the efficacy of a given treatment regimen. Detection can be facilitated by coupling (*i.e.*, physically linking) the antibody to a detectable substance. Examples of detectable substances include various enzymes, prosthetic groups, fluorescent materials, luminescent materials, bioluminescent materials, and radioactive materials. Examples of suitable enzymes include horseradish peroxidase, alkaline phosphatase, β-galactosidase, or acetylcholinesterase; examples of suitable prosthetic group complexes include streptavidin/biotin and avidin/biotin; examples of suitable fluorescent materials include umbelliferone, fluorescein, fluorescein isothiocyanate, rhodamine, dichlorotriazinylamine fluorescein, dansyl chloride or phycoerythrin; an example of a luminescent material includes luminol; examples of bioluminescent materials include luciferase, luciferin, and aequorin, and examples of suitable radioactive material include ^{125}I, ^{131}I, ^{35}S or ^{3}H.

NOVX Recombinant Expression Vectors and Host Cells

Another aspect of the invention pertains to vectors, preferably expression vectors, containing a nucleic acid encoding an NOVX protein, or derivatives, fragments, analogs or homologs thereof. As used herein, the term "vector" refers to a nucleic acid molecule capable of transporting another nucleic acid to which it has been linked. One type of vector is a "plasmid", which refers to a circular double stranded DNA loop into which additional DNA segments can be ligated. Another type of vector is a viral vector, wherein additional DNA segments can be ligated into the viral genome. Certain vectors are capable of autonomous replication in a host cell into which they are introduced (*e.g.*, bacterial vectors having a bacterial origin of replication and episomal mammalian vectors). Other vectors (*e.g.*, non-episomal mammalian vectors) are integrated into the genome of a host cell upon introduction into the host cell, and thereby are replicated along with the host genome. Moreover, certain vectors are capable of directing the expression of genes to which they are operatively-linked. Such vectors are referred to herein as "expression vectors". In general, expression vectors of utility in recombinant DNA techniques are often in the form of plasmids. In the present specification, "plasmid" and "vector" can be used interchangeably as the plasmid is the most commonly used form of vector. However, the invention is intended to include such other forms of expression vectors, such as viral vectors (*e.g.*, replication defective retroviruses, adenoviruses and adeno-associated viruses), which serve equivalent functions.

The recombinant expression vectors of the invention comprise a nucleic acid of the invention in a form suitable for expression of the nucleic acid in a host cell, which means that the recombinant expression vectors include one or more regulatory sequences, selected on the basis of the host cells to be used for expression, that is operatively-linked to the nucleic acid sequence to be expressed. Within a recombinant expression vector, "operably-linked" is intended to mean that the nucleotide sequence of interest is linked to the regulatory sequence(s) in a manner that allows for expression of the nucleotide sequence (*e.g.*, in an *in vitro* transcription/translation system or in a host cell when the vector is introduced into the host cell).

The term "regulatory sequence" is intended to includes promoters, enhancers and other expression control elements (*e.g.*, polyadenylation signals). Such regulatory sequences are described, for example, in Goeddel, GENE EXPRESSION TECHNOLOGY: METHODS IN ENZYMOLOGY 185, Academic Press, San Diego, Calif. (1990). Regulatory sequences include those that direct constitutive expression of a nucleotide sequence in many types of host cell and those that direct expression of the nucleotide sequence only in certain host cells (*e.g.*, tissue-specific regulatory sequences). It will be appreciated by those skilled in the art that the design of the expression vector can depend on such factors as the choice of the host cell to be

transformed, the level of expression of protein desired, etc. The expression vectors of the invention can be introduced into host cells to thereby produce proteins or peptides, including fusion proteins or peptides, encoded by nucleic acids as described herein (*e.g.*, NOVX proteins, mutant forms of NOVX proteins, fusion proteins, etc.).

The recombinant expression vectors of the invention can be designed for expression of NOVX proteins in prokaryotic or eukaryotic cells. For example, NOVX proteins can be expressed in bacterial cells such as *Escherichia coli*, insect cells (using baculovirus expression vectors) yeast cells or mammalian cells. Suitable host cells are discussed further in Goeddel, GENE EXPRESSION TECHNOLOGY: METHODS IN ENZYMOLOGY 185, Academic Press, San Diego, Calif. (1990). Alternatively, the recombinant expression vector can be transcribed and translated *in vitro*, for example using T7 promoter regulatory sequences and T7 polymerase.

Expression of proteins in prokaryotes is most often carried out in *Escherichia coli* with vectors containing constitutive or inducible promoters directing the expression of either fusion or non-fusion proteins. Fusion vectors add a number of amino acids to a protein encoded therein, usually to the amino terminus of the recombinant protein. Such fusion vectors typically serve three purposes: (*i*) to increase expression of recombinant protein; (*ii*) to increase the solubility of the recombinant protein; and (*iii*) to aid in the purification of the recombinant protein by acting as a ligand in affinity purification. Often, in fusion expression vectors, a proteolytic cleavage site is introduced at the junction of the fusion moiety and the recombinant protein to enable separation of the recombinant protein from the fusion moiety subsequent to purification of the fusion protein. Such enzymes, and their cognate recognition sequences, include Factor Xa, thrombin and enterokinase. Typical fusion expression vectors include pGEX (Pharmacia Biotech Inc; Smith and Johnson, 1988. *Gene* 67: 31-40), pMAL (New England Biolabs, Beverly, Mass.) and pRIT5 (Pharmacia, Piscataway, N.J.) that fuse glutathione S-transferase (GST), maltose E binding protein, or protein A, respectively, to the target recombinant protein.

Examples of suitable inducible non-fusion *E. coli* expression vectors include pTrc (Amrann *et al.*, (1988) *Gene* 69:301-315) and pET 11d (Studier *et al.*, GENE EXPRESSION TECHNOLOGY: METHODS IN ENZYMOLOGY 185, Academic Press, San Diego, Calif. (1990) 60-89).

One strategy to maximize recombinant protein expression in *E. coli* is to express the protein in a host bacteria with an impaired capacity to proteolytically cleave the recombinant protein. *See, e.g.,* Gottesman, GENE EXPRESSION TECHNOLOGY: METHODS IN ENZYMOLOGY 185, Academic Press, San Diego, Calif. (1990) 119-128. Another strategy is to alter the nucleic acid sequence of the nucleic acid to be inserted into an expression vector so that the individual codons for each amino acid are those preferentially utilized in *E. coli* (*see, e.g.,* Wada, *et al.*, 1992. *Nucl.*

Acids Res. 20: 2111-2118). Such alteration of nucleic acid sequences of the invention can be carried out by standard DNA synthesis techniques.

In another embodiment, the NOVX expression vector is a yeast expression vector. Examples of vectors for expression in yeast *Saccharomyces cerivisae* include pYepSec1 (Baldari, *et al.*, 1987. *EMBO J.* 6: 229-234), pMFa (Kurjan and Herskowitz, 1982. *Cell* 30: 933-943), pJRY88 (Schultz *et al.*, 1987. *Gene* 54: 113-123), pYES2 (Invitrogen Corporation, San Diego, Calif.), and picZ (InVitrogen Corp, San Diego, Calif.).

Alternatively, NOVX can be expressed in insect cells using baculovirus expression vectors. Baculovirus vectors available for expression of proteins in cultured insect cells (*e.g.*, SF9 cells) include the pAc series (Smith, *et al.*, 1983. *Mol. Cell. Biol.* 3: 2156-2165) and the pVL series (Lucklow and Summers. 1989. *Virology* 170: 31-39).

In yet another embodiment, a nucleic acid of the invention is expressed in mammalian cells using a mammalian expression vector. Examples of mammalian expression vectors include pCDM8 (Seed, 1987. *Nature* 329: 840) and pMT2PC (Kaufman, *et al.*, 1987. *EMBO J.* 6: 187-195). When used in mammalian cells, the expression vector's control functions are often provided by viral regulatory elements. For example, commonly used promoters are derived from polyoma, adenovirus 2, cytomegalovirus, and simian virus 40. For other suitable expression systems for both prokaryotic and eukaryotic cells see, *e.g.*, Chapters 16 and 17 of Sambrook, *et al.*, MOLECULAR CLONING: A LABORATORY MANUAL. 2nd ed., Cold Spring Harbor Laboratory, Cold Spring Harbor Laboratory Press. Cold Spring Harbor, N.Y., 1989.

In another embodiment, the recombinant mammalian expression vector is capable of directing expression of the nucleic acid preferentially in a particular cell type (*e.g.*, tissue-specific regulatory elements are used to express the nucleic acid). Tissue-specific regulatory elements are known in the art. Non-limiting examples of suitable tissue-specific promoters include the albumin promoter (liver-specific; Pinkert, *et al.*, 1987. *Genes Dev.* 1: 268-277), lymphoid-specific promoters (Calame and Eaton, 1988. *Adv. Immunol.* 43: 235-275), in particular promoters of T cell receptors (Winoto and Baltimore, 1989. *EMBO J.* 8: 729-733) and immunoglobulins (Banerji, *et al.*, 1983. *Cell* 33: 729-740; Queen and Baltimore, 1983. *Cell* 33: 741-748), neuron-specific promoters (*e.g.*, the neurofilament promoter; Byrne and Ruddle, 1989. *Proc. Natl. Acad. Sci. USA* 86: 5473-5477), pancreas-specific promoters (Edlund, *et al.*, 1985. *Science* 230: 912-916), and mammary gland-specific promoters (*e.g.*, milk whey promoter; U.S. Pat. No. 4,873,316 and European Application Publication No. 264,166). Developmentally-regulated promoters are also encompassed, *e.g.*, the murine hox promoters (Kessel and Gruss, 1990. *Science* 249: 374-379) and the α-fetoprotein promoter (Campes and Tilghman, 1989. *Genes Dev.* 3: 537-546).

The invention further provides a recombinant expression vector comprising a DNA molecule of the invention cloned into the expression vector in an antisense orientation. That is, the DNA molecule is operatively-linked to a regulatory sequence in a manner that allows for expression (by transcription of the DNA molecule) of an RNA molecule that is antisense to NOVX mRNA. Regulatory sequences operatively linked to a nucleic acid cloned in the antisense orientation can be chosen that direct the continuous expression of the antisense RNA molecule in a variety of cell types, for instance viral promoters and/or enhancers, or regulatory sequences can be chosen that direct constitutive, tissue specific or cell type specific expression of antisense RNA. The antisense expression vector can be in the form of a recombinant plasmid, phagemid or attenuated virus in which antisense nucleic acids are produced under the control of a high efficiency regulatory region, the activity of which can be determined by the cell type into which the vector is introduced. For a discussion of the regulation of gene expression using antisense genes *see, e.g.,* Weintraub, *et al.*, "Antisense RNA as a molecular tool for genetic analysis," *Reviews-Trends in Genetics*, Vol. 1(1) 1986.

Another aspect of the invention pertains to host cells into which a recombinant expression vector of the invention has been introduced. The terms "host cell" and "recombinant host cell" are used interchangeably herein. It is understood that such terms refer not only to the particular subject cell but also to the progeny or potential progeny of such a cell. Because certain modifications may occur in succeeding generations due to either mutation or environmental influences, such progeny may not, in fact, be identical to the parent cell, but are still included within the scope of the term as used herein.

A host cell can be any prokaryotic or eukaryotic cell. For example, NOVX protein can be expressed in bacterial cells such as *E. coli*, insect cells, yeast or mammalian cells (such as Chinese hamster ovary cells (CHO) or COS cells). Other suitable host cells are known to those skilled in the art.

Vector DNA can be introduced into prokaryotic or eukaryotic cells via conventional transformation or transfection techniques. As used herein, the terms "transformation" and "transfection" are intended to refer to a variety of art-recognized techniques for introducing foreign nucleic acid (*e.g.*, DNA) into a host cell, including calcium phosphate or calcium chloride co-precipitation, DEAE-dextran-mediated transfection, lipofection, or electroporation. Suitable methods for transforming or transfecting host cells can be found in Sambrook, *et al.* (MOLECULAR CLONING: A LABORATORY MANUAL. 2nd ed., Cold Spring Harbor Laboratory, Cold Spring Harbor Laboratory Press, Cold Spring Harbor, N.Y., 1989), and other laboratory manuals.

For stable transfection of mammalian cells, it is known that, depending upon the expression vector and transfection technique used, only a small fraction of cells may integrate the foreign DNA into their genome. In order to identify and select these integrants, a gene that encodes a selectable marker (*e.g.*, resistance to antibiotics) is generally introduced into the host cells along with the gene of interest. Various selectable markers include those that confer resistance to drugs, such as G418, hygromycin and methotrexate. Nucleic acid encoding a selectable marker can be introduced into a host cell on the same vector as that encoding NOVX or can be introduced on a separate vector. Cells stably transfected with the introduced nucleic acid can be identified by drug selection (*e.g.*, cells that have incorporated the selectable marker gene will survive, while the other cells die).

A host cell of the invention, such as a prokaryotic or eukaryotic host cell in culture, can be used to produce (*i.e.*, express) NOVX protein. Accordingly, the invention further provides methods for producing NOVX protein using the host cells of the invention. In one embodiment, the method comprises culturing the host cell of invention (into which a recombinant expression vector encoding NOVX protein has been introduced) in a suitable medium such that NOVX protein is produced. In another embodiment, the method further comprises isolating NOVX protein from the medium or the host cell.

Transgenic NOVX Animals

The host cells of the invention can also be used to produce non-human transgenic animals. For example, in one embodiment, a host cell of the invention is a fertilized oocyte or an embryonic stem cell into which NOVX protein-coding sequences have been introduced. Such host cells can then be used to create non-human transgenic animals in which exogenous NOVX sequences have been introduced into their genome or homologous recombinant animals in which endogenous NOVX sequences have been altered. Such animals are useful for studying the function and/or activity of NOVX protein and for identifying and/or evaluating modulators of NOVX protein activity. As used herein, a "transgenic animal" is a non-human animal, preferably a mammal, more preferably a rodent such as a rat or mouse, in which one or more of the cells of the animal includes a transgene. Other examples of transgenic animals include non-human primates, sheep, dogs, cows, goats, chickens, amphibians, etc. A transgene is exogenous DNA that is integrated into the genome of a cell from which a transgenic animal develops and that remains in the genome of the mature animal, thereby directing the expression of an encoded gene product in one or more cell types or tissues of the transgenic animal. As used herein, a "homologous recombinant animal" is a non-human animal, preferably a mammal, more preferably a mouse, in which an endogenous NOVX gene has been altered by homologous

recombination between the endogenous gene and an exogenous DNA molecule introduced into a cell of the animal, *e.g.*, an embryonic cell of the animal, prior to development of the animal.

A transgenic animal of the invention can be created by introducing NOVX-encoding nucleic acid into the male pronuclei of a fertilized oocyte (*e.g.*, by microinjection, retroviral infection) and allowing the oocyte to develop in a pseudopregnant female foster animal. The human NOVX cDNA sequences SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102 can be introduced as a transgene into the genome of a non-human animal. Alternatively, a non-human homologue of the human NOVX gene, such as a mouse NOVX gene, can be isolated based on hybridization to the human NOVX cDNA (described further *supra*) and used as a transgene. Intronic sequences and polyadenylation signals can also be included in the transgene to increase the efficiency of expression of the transgene. A tissue-specific regulatory sequence(s) can be operably-linked to the NOVX transgene to direct expression of NOVX protein to particular cells. Methods for generating transgenic animals via embryo manipulation and microinjection, particularly animals such as mice, have become conventional in the art and are described, for example, in U.S. Patent Nos. 4,736,866; 4,870,009; and 4,873,191; and Hogan, 1986. In: MANIPULATING THE MOUSE EMBRYO, Cold Spring Harbor Laboratory Press, Cold Spring Harbor, N.Y. Similar methods are used for production of other transgenic animals. A transgenic founder animal can be identified based upon the presence of the NOVX transgene in its genome and/or expression of NOVX mRNA in tissues or cells of the animals. A transgenic founder animal can then be used to breed additional animals carrying the transgene. Moreover, transgenic animals carrying a transgene-encoding NOVX protein can further be bred to other transgenic animals carrying other transgenes.

To create a homologous recombinant animal, a vector is prepared which contains at least a portion of an NOVX gene into which a deletion, addition or substitution has been introduced to thereby alter, *e.g.*, functionally disrupt, the NOVX gene. The NOVX gene can be a human gene (*e.g.*, the cDNA of SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102), but more preferably, is a non-human homologue of a human NOVX gene. For example, a mouse homologue of human NOVX gene of SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102 can be used to construct a homologous recombination vector suitable for altering an endogenous NOVX gene in the mouse genome. In one embodiment, the vector is designed such that, upon homologous recombination, the endogenous NOVX gene is functionally disrupted (*i.e.*, no longer encodes a functional protein; also referred to as a "knock out" vector).

Alternatively, the vector can be designed such that, upon homologous recombination, the endogenous NOVX gene is mutated or otherwise altered but still encodes functional protein

(*e.g.*, the upstream regulatory region can be altered to thereby alter the expression of the endogenous NOVX protein). In the homologous recombination vector, the altered portion of the NOVX gene is flanked at its 5'- and 3'-termini by additional nucleic acid of the NOVX gene to allow for homologous recombination to occur between the exogenous NOVX gene carried by the vector and an endogenous NOVX gene in an embryonic stem cell. The additional flanking NOVX nucleic acid is of sufficient length for successful homologous recombination with the endogenous gene. Typically, several kilobases of flanking DNA (both at the 5'- and 3'-termini) are included in the vector. *See, e.g.,* Thomas, *et al.*, 1987. *Cell* 51: 503 for a description of homologous recombination vectors. The vector is ten introduced into an embryonic stem cell line (*e.g.*, by electroporation) and cells in which the introduced NOVX gene has homologously-recombined with the endogenous NOVX gene are selected. *See, e.g.*, Li, *et al.*, 1992. *Cell* 69: 915.

The selected cells are then injected into a blastocyst of an animal (*e.g.*, a mouse) to form aggregation chimeras. *See, e.g.*, Bradley, 1987. In: TERATOCARCINOMAS AND EMBRYONIC STEM CELLS: A PRACTICAL APPROACH, Robertson, ed. IRL, Oxford, pp. 113-152. A chimeric embryo can then be implanted into a suitable pseudopregnant female foster animal and the embryo brought to term. Progeny harboring the homologously-recombined DNA in their germ cells can be used to breed animals in which all cells of the animal contain the homologously-recombined DNA by germline transmission of the transgene. Methods for constructing homologous recombination vectors and homologous recombinant animals are described further in Bradley, 1991. *Curr. Opin. Biotechnol.* 2: 823-829; PCT International Publication Nos.: WO 90/11354; WO 91/01140; WO 92/0968; and WO 93/04169.

In another embodiment, transgenic non-humans animals can be produced that contain selected systems that allow for regulated expression of the transgene. One example of such a system is the cre/loxP recombinase system of bacteriophage P1. For a description of the cre/loxP recombinase system, *See, e.g.*, Lakso, *et al.*, 1992. *Proc. Natl. Acad. Sci. USA* 89: 6232-6236. Another example of a recombinase system is the FLP recombinase system of *Saccharomyces cerevisiae. See,* O'Gorman, *et al.*, 1991. *Science* 251:1351-1355. If a cre/loxP recombinase system is used to regulate expression of the transgene, animals containing transgenes encoding both the Cre recombinase and a selected protein are required. Such animals can be provided through the construction of "double" transgenic animals, *e.g.*, by mating two transgenic animals, one containing a transgene encoding a selected protein and the other containing a transgene encoding a recombinase.

Clones of the non-human transgenic animals described herein can also be produced according to the methods described in Wilmut, *et al.*, 1997. *Nature* 385: 810-813. In brief, a cell

(*e.g.*, a somatic cell) from the transgenic animal can be isolated and induced to exit the growth cycle and enter G_0 phase. The quiescent cell can then be fused, *e.g.*, through the use of electrical pulses, to an enucleated oocyte from an animal of the same species from which the quiescent cell is isolated. The reconstructed oocyte is then cultured such that it develops to morula or blastocyte and then transferred to pseudopregnant female foster animal. The offspring borne of this female foster animal will be a clone of the animal from which the cell (*e.g.*, the somatic cell) is isolated.

Pharmaceutical Compositions

The NOVX nucleic acid molecules, NOVX proteins, and anti-NOVX antibodies (also referred to herein as "active compounds") of the invention, and derivatives, fragments, analogs and homologs thereof, can be incorporated into pharmaceutical compositions suitable for administration. Such compositions typically comprise the nucleic acid molecule, protein, or antibody and a pharmaceutically acceptable carrier. As used herein, "pharmaceutically acceptable carrier" is intended to include any and all solvents, dispersion media, coatings, antibacterial and antifungal agents, isotonic and absorption delaying agents, and the like, compatible with pharmaceutical administration. Suitable carriers are described in the most recent edition of Remington's Pharmaceutical Sciences, a standard reference text in the field, which is incorporated herein by reference. Preferred examples of such carriers or diluents include, but are not limited to, water, saline, finger's solutions, dextrose solution, and 5% human serum albumin. Liposomes and non-aqueous vehicles such as fixed oils may also be used. The use of such media and agents for pharmaceutically active substances is well known in the art. Except insofar as any conventional media or agent is incompatible with the active compound, use thereof in the compositions is contemplated. Supplementary active compounds can also be incorporated into the compositions.

A pharmaceutical composition of the invention is formulated to be compatible with its intended route of administration. Examples of routes of administration include parenteral, *e.g.*, intravenous, intradermal, subcutaneous, oral (*e.g.*, inhalation), transdermal (*i.e.*, topical), transmucosal, and rectal administration. Solutions or suspensions used for parenteral, intradermal, or subcutaneous application can include the following components: a sterile diluent such as water for injection, saline solution, fixed oils, polyethylene glycols, glycerine, propylene glycol or other synthetic solvents; antibacterial agents such as benzyl alcohol or methyl parabens; antioxidants such as ascorbic acid or sodium bisulfite; chelating agents such as ethylenediaminetetraacetic acid (EDTA); buffers such as acetates, citrates or phosphates, and agents for the adjustment of tonicity such as sodium chloride or dextrose. The pH can be

adjusted with acids or bases, such as hydrochloric acid or sodium hydroxide. The parenteral preparation can be enclosed in ampoules, disposable syringes or multiple dose vials made of glass or plastic.

Pharmaceutical compositions suitable for injectable use include sterile aqueous solutions (where water soluble) or dispersions and sterile powders for the extemporaneous preparation of sterile injectable solutions or dispersion. For intravenous administration, suitable carriers include physiological saline, bacteriostatic water, Cremophor EL™ (BASF, Parsippany, N.J.) or phosphate buffered saline (PBS). In all cases, the composition must be sterile and should be fluid to the extent that easy syringeability exists. It must be stable under the conditions of manufacture and storage and must be preserved against the contaminating action of microorganisms such as bacteria and fungi. The carrier can be a solvent or dispersion medium containing, for example, water, ethanol, polyol (for example, glycerol, propylene glycol, and liquid polyethylene glycol, and the like), and suitable mixtures thereof. The proper fluidity can be maintained, for example, by the use of a coating such as lecithin, by the maintenance of the required particle size in the case of dispersion and by the use of surfactants. Prevention of the action of microorganisms can be achieved by various antibacterial and antifungal agents, for example, parabens, chlorobutanol, phenol, ascorbic acid, thimerosal, and the like. In many cases, it will be preferable to include isotonic agents, for example, sugars, polyalcohols such as manitol, sorbitol, sodium chloride in the composition. Prolonged absorption of the injectable compositions can be brought about by including in the composition an agent which delays absorption, for example, aluminum monostearate and gelatin.

Sterile injectable solutions can be prepared by incorporating the active compound (*e.g.*, an NOVX protein or anti-NOVX antibody) in the required amount in an appropriate solvent with one or a combination of ingredients enumerated above, as required, followed by filtered sterilization. Generally, dispersions are prepared by incorporating the active compound into a sterile vehicle that contains a basic dispersion medium and the required other ingredients from those enumerated above. In the case of sterile powders for the preparation of sterile injectable solutions, methods of preparation are vacuum drying and freeze-drying that yields a powder of the active ingredient plus any additional desired ingredient from a previously sterile-filtered solution thereof.

Oral compositions generally include an inert diluent or an edible carrier. They can be enclosed in gelatin capsules or compressed into tablets. For the purpose of oral therapeutic administration, the active compound can be incorporated with excipients and used in the form of tablets, troches, or capsules. Oral compositions can also be prepared using a fluid carrier for use as a mouthwash, wherein the compound in the fluid carrier is applied orally and swished and

expectorated or swallowed. Pharmaceutically compatible binding agents, and/or adjuvant materials can be included as part of the composition. The tablets, pills, capsules, troches and the like can contain any of the following ingredients, or compounds of a similar nature: a binder such as microcrystalline cellulose, gum tragacanth or gelatin; an excipient such as starch or lactose, a disintegrating agent such as alginic acid, Primogel, or corn starch; a lubricant such as magnesium stearate or Sterotes; a glidant such as colloidal silicon dioxide; a sweetening agent such as sucrose or saccharin; or a flavoring agent such as peppermint. methyl salicylate, or orange flavoring.

For administration by inhalation, the compounds are delivered in the form of an aerosol spray from pressured container or dispenser which contains a suitable propellant, *e.g.*, a gas such as carbon dioxide, or a nebulizer.

Systemic administration can also be by transmucosal or transdermal means. For transmucosal or transdermal administration, penetrants appropriate to the barrier to be permeated are used in the formulation. Such penetrants are generally known in the art, and include, for example, for transmucosal administration, detergents, bile salts, and fusidic acid derivatives. Transmucosal administration can be accomplished through the use of nasal sprays or suppositories. For transdermal administration, the active compounds are formulated into ointments, salves, gels, or creams as generally known in the art.

The compounds can also be prepared in the form of suppositories (*e.g.*, with conventional suppository bases such as cocoa butter and other glycerides) or retention enemas for rectal delivery.

In one embodiment, the active compounds are prepared with carriers that will protect the compound against rapid elimination from the body, such as a controlled release formulation, including implants and microencapsulated delivery systems. Biodegradable, biocompatible polymers can be used, such as ethylene vinyl acetate, polyanhydrides, polyglycolic acid, collagen, polyorthoesters, and polylactic acid. Methods for preparation of such formulations will be apparent to those skilled in the art. The materials can also be obtained commercially from Alza Corporation and Nova Pharmaceuticals, Inc. Liposomal suspensions (including liposomes targeted to infected cells with monoclonal antibodies to viral antigens) can also be used as pharmaceutically acceptable carriers. These can be prepared according to methods known to those skilled in the art, for example, as described in U.S. Patent No. 4,522,811.

It is especially advantageous to formulate oral or parenteral compositions in dosage unit form for ease of administration and uniformity of dosage. Dosage unit form as used herein refers to physically discrete units suited as unitary dosages for the subject to be treated; each unit containing a predetermined quantity of active compound calculated to produce the desired

therapeutic effect in association with the required pharmaceutical carrier. The specification for the dosage unit forms of the invention are dictated by and directly dependent on the unique characteristics of the active compound and the particular therapeutic effect to be achieved, and the limitations inherent in the art of compounding such an active compound for the treatment of individuals.

The nucleic acid molecules of the invention can be inserted into vectors and used as gene therapy vectors. Gene therapy vectors can be delivered to a subject by, for example, intravenous injection, local administration (*see, e.g.*, U.S. Patent No. 5,328,470) or by stereotactic injection (*see, e.g.*, Chen, *et al.*, 1994. *Proc. Natl. Acad. Sci. USA* 91: 3054-3057). The pharmaceutical preparation of the gene therapy vector can include the gene therapy vector in an acceptable diluent, or can comprise a slow release matrix in which the gene delivery vehicle is imbedded. Alternatively, where the complete gene delivery vector can be produced intact from recombinant cells, *e.g.*, retroviral vectors, the pharmaceutical preparation can include one or more cells that produce the gene delivery system.

The pharmaceutical compositions can be included in a container, pack, or dispenser together with instructions for administration.

Screening and Detection Methods

The isolated nucleic acid molecules of the invention can be used to express NOVX protein (*e.g.*, via a recombinant expression vector in a host cell in gene therapy applications), to detect NOVX mRNA (*e.g.*, in a biological sample) or a genetic lesion in an NOVX gene, and to modulate NOVX activity, as described further, below. In addition, the NOVX proteins can be used to screen drugs or compounds that modulate the NOVX protein activity or expression as well as to treat disorders characterized by insufficient or excessive production of NOVX protein or production of NOVX protein forms that have decreased or aberrant activity compared to NOVX wild-type protein (*e.g.*; diabetes (regulates insulin release); obesity (binds and transport lipids); metabolic disturbances associated with obesity, the metabolic syndrome X as well as anorexia and wasting disorders associated with chronic diseases and various cancers, and infectious disease(possesses anti-microbial activity) and the various dyslipidemias. In addition, the anti-NOVX antibodies of the invention can be used to detect and isolate NOVX proteins and modulate NOVX activity. In yet a further aspect, the invention can be used in methods to influence appetite, absorption of nutrients and the disposition of metabolic substrates in both a positive and negative fashion.

The invention further pertains to novel agents identified by the screening assays described herein and uses thereof for treatments as described, *supra*.

Screening Assays

The invention provides a method (also referred to herein as a "screening assay") for identifying modulators, *i.e.*, candidate or test compounds or agents (*e.g.*, peptides, peptidomimetics, small molecules or other drugs) that bind to NOVX proteins or have a stimulatory or inhibitory effect on, *e.g.*, NOVX protein expression or NOVX protein activity. The invention also includes compounds identified in the screening assays described herein.

In one embodiment, the invention provides assays for screening candidate or test compounds which bind to or modulate the activity of the membrane-bound form of an NOVX protein or polypeptide or biologically-active portion thereof. The test compounds of the invention can be obtained using any of the numerous approaches in combinatorial library methods known in the art, including: biological libraries; spatially addressable parallel solid phase or solution phase libraries; synthetic library methods requiring deconvolution; the "one-bead one-compound" library method; and synthetic library methods using affinity chromatography selection. The biological library approach is limited to peptide libraries, while the other four approaches are applicable to peptide, non-peptide oligomer or small molecule libraries of compounds. *See, e.g.*, Lam, 1997. *Anticancer Drug Design* 12: 145.

A "small molecule" as used herein, is meant to refer to a composition that has a molecular weight of less than about 5 kD and most preferably less than about 4 kD. Small molecules can be, *e.g.*, nucleic acids, peptides, polypeptides, peptidomimetics, carbohydrates, lipids or other organic or inorganic molecules. Libraries of chemical and/or biological mixtures, such as fungal, bacterial, or algal extracts, are known in the art and can be screened with any of the assays of the invention.

Examples of methods for the synthesis of molecular libraries can be found in the art, for example in: DeWitt, *et al.*, 1993. *Proc. Natl. Acad. Sci. U.S.A.* 90: 6909; Erb, *et al.*, 1994. *Proc. Natl. Acad. Sci. U.S.A.* 91: 11422; Zuckermann, *et al.*, 1994. *J. Med. Chem.* 37: 2678; Cho, *et al.*, 1993. *Science* 261: 1303; Carrell, *et al.*, 1994. *Angew. Chem. Int. Ed. Engl.* 33: 2059; Carell, *et al.*, 1994. *Angew. Chem. Int. Ed. Engl.* 33: 2061; and Gallop, *et al.*, 1994. *J. Med. Chem.* 37: 1233.

Libraries of compounds may be presented in solution (*e.g.*, Houghten, 1992. *Biotechniques* 13: 412-421), or on beads (Lam, 1991. *Nature* 354: 82-84), on chips (Fodor, 1993. *Nature* 364: 555-556), bacteria (Ladner, U.S. Patent No. 5,223,409), spores (Ladner, U.S. Patent 5,233,409), plasmids (Cull, *et al.*, 1992. *Proc. Natl. Acad. Sci. USA* 89: 1865-1869) or on phage (Scott and Smith, 1990. *Science* 249: 386-390; Devlin, 1990. *Science* 249: 404-406; Cwirla, *et*

al., 1990. *Proc. Natl Acad. Sci. U.S.A.* 87: 6378-6382; Felici, 1991. *J. Mol. Biol.* 222: 301-310; Ladner, U.S. Patent No. 5,233,409.).

In one embodiment, an assay is a cell-based assay in which a cell which expresses a membrane-bound form of NOVX protein, or a biologically-active portion thereof, on the cell surface is contacted with a test compound and the ability of the test compound to bind to an NOVX protein determined. The cell, for example, can of mammalian origin or a yeast cell. Determining the ability of the test compound to bind to the NOVX protein can be accomplished, for example, by coupling the test compound with a radioisotope or enzymatic label such that binding of the test compound to the NOVX protein or biologically-active portion thereof can be determined by detecting the labeled compound in a complex. For example, test compounds can be labeled with ^{125}I, ^{35}S, ^{14}C, or ^{3}H, either directly or indirectly, and the radioisotope detected by direct counting of radioemission or by scintillation counting. Alternatively, test compounds can be enzymatically-labeled with, for example, horseradish peroxidase, alkaline phosphatase, or luciferase, and the enzymatic label detected by determination of conversion of an appropriate substrate to product. In one embodiment, the assay comprises contacting a cell which expresses a membrane-bound form of NOVX protein, or a biologically-active portion thereof, on the cell surface with a known compound which binds NOVX to form an assay mixture, contacting the assay mixture with a test compound, and determining the ability of the test compound to interact with an NOVX protein, wherein determining the ability of the test compound to interact with an NOVX protein comprises determining the ability of the test compound to preferentially bind to NOVX protein or a biologically-active portion thereof as compared to the known compound.

In another embodiment, an assay is a cell-based assay comprising contacting a cell expressing a membrane-bound form of NOVX protein, or a biologically-active portion thereof, on the cell surface with a test compound and determining the ability of the test compound to modulate (*e.g.*, stimulate or inhibit) the activity of the NOVX protein or biologically-active portion thereof. Determining the ability of the test compound to modulate the activity of NOVX or a biologically-active portion thereof can be accomplished, for example, by determining the ability of the NOVX protein to bind to or interact with an NOVX target molecule. As used herein, a "target molecule" is a molecule with which an NOVX protein binds or interacts in nature, for example, a molecule on the surface of a cell which expresses an NOVX interacting protein, a molecule on the surface of a second cell, a molecule in the extracellular milieu, a molecule associated with the internal surface of a cell membrane or a cytoplasmic molecule. An NOVX target molecule can be a non-NOVX molecule or an NOVX protein or polypeptide of the invention. In one embodiment, an NOVX target molecule is a component of a signal transduction pathway that facilitates transduction of an extracellular signal (*e.g.* a signal

generated by binding of a compound to a membrane-bound NOVX molecule) through the cell membrane and into the cell. The target, for example, can be a second intercellular protein that has catalytic activity or a protein that facilitates the association of downstream signaling molecules with NOVX.

Determining the ability of the NOVX protein to bind to or interact with an NOVX target molecule can be accomplished by one of the methods described above for determining direct binding. In one embodiment, determining the ability of the NOVX protein to bind to or interact with an NOVX target molecule can be accomplished by determining the activity of the target molecule. For example, the activity of the target molecule can be determined by detecting induction of a cellular second messenger of the target (*i.e.* intracellular Ca^{2+}, diacylglycerol, IP_3, etc.), detecting catalytic/enzymatic activity of the target an appropriate substrate, detecting the induction of a reporter gene (comprising an NOVX-responsive regulatory element operatively linked to a nucleic acid encoding a detectable marker, *e.g.*, luciferase), or detecting a cellular response, for example, cell survival, cellular differentiation, or cell proliferation.

In yet another embodiment, an assay of the invention is a cell-free assay comprising contacting an NOVX protein or biologically-active portion thereof with a test compound and determining the ability of the test compound to bind to the NOVX protein or biologically-active portion thereof. Binding of the test compound to the NOVX protein can be determined either directly or indirectly as described above. In one such embodiment, the assay comprises contacting the NOVX protein or biologically-active portion thereof with a known compound which binds NOVX to form an assay mixture, contacting the assay mixture with a test compound, and determining the ability of the test compound to interact with an NOVX protein, wherein determining the ability of the test compound to interact with an NOVX protein comprises determining the ability of the test compound to preferentially bind to NOVX or biologically-active portion thereof as compared to the known compound.

In still another embodiment, an assay is a cell-free assay comprising contacting NOVX protein or biologically-active portion thereof with a test compound and determining the ability of the test compound to modulate (*e.g.* stimulate or inhibit) the activity of the NOVX protein or biologically-active portion thereof. Determining the ability of the test compound to modulate the activity of NOVX can be accomplished, for example, by determining the ability of the NOVX protein to bind to an NOVX target molecule by one of the methods described above for determining direct binding. In an alternative embodiment, determining the ability of the test compound to modulate the activity of NOVX protein can be accomplished by determining the ability of the NOVX protein further modulate an NOVX target molecule. For example, the

catalytic/enzymatic activity of the target molecule on an appropriate substrate can be determined as described, *supra*.

In yet another embodiment, the cell-free assay comprises contacting the NOVX protein or biologically-active portion thereof with a known compound which binds NOVX protein to form an assay mixture, contacting the assay mixture with a test compound, and determining the ability of the test compound to interact with an NOVX protein, wherein determining the ability of the test compound to interact with an NOVX protein comprises determining the ability of the NOVX protein to preferentially bind to or modulate the activity of an NOVX target molecule.

The cell-free assays of the invention are amenable to use of both the soluble form or the membrane-bound form of NOVX protein. In the case of cell-free assays comprising the membrane-bound form of NOVX protein, it may be desirable to utilize a solubilizing agent such that the membrane-bound form of NOVX protein is maintained in solution. Examples of such solubilizing agents include non-ionic detergents such as n-octylglucoside, n-dodecylglucoside, n-dodecylmaltoside, octanoyl-N-methylglucamide, decanoyl-N-methylglucamide, Triton® X-100, Triton® X-114, Thesit®, Isotridecypoly(ethylene glycol ether)$_n$, N-dodecyl--N,N-dimethyl-3-ammonio-1-propane sulfonate, 3-(3-cholamidopropyl) dimethylamminiol-1-propane sulfonate (CHAPS), or 3-(3-cholamidopropyl)dimethylamminiol-2-hydroxy-1-propane sulfonate (CHAPSO).

In more than one embodiment of the above assay methods of the invention, it may be desirable to immobilize either NOVX protein or its target molecule to facilitate separation of complexed from uncomplexed forms of one or both of the proteins, as well as to accommodate automation of the assay. Binding of a test compound to NOVX protein, or interaction of NOVX protein with a target molecule in the presence and absence of a candidate compound, can be accomplished in any vessel suitable for containing the reactants. Examples of such vessels include microtiter plates, test tubes, and micro-centrifuge tubes. In one embodiment, a fusion protein can be provided that adds a domain that allows one or both of the proteins to be bound to a matrix. For example, GST-NOVX fusion proteins or GST-target fusion proteins can be adsorbed onto glutathione sepharose beads (Sigma Chemical, St. Louis, MO) or glutathione derivatized microtiter plates, that are then combined with the test compound or the test compound and either the non-adsorbed target protein or NOVX protein, and the mixture is incubated under conditions conducive to complex formation (*e.g.*, at physiological conditions for salt and pH). Following incubation, the beads or microtiter plate wells are washed to remove any unbound components, the matrix immobilized in the case of beads, complex determined either directly or indirectly, for example, as described, *supra*. Alternatively, the complexes can

be dissociated from the matrix, and the level of NOVX protein binding or activity determined using standard techniques.

Other techniques for immobilizing proteins on matrices can also be used in the screening assays of the invention. For example, either the NOVX protein or its target molecule can be immobilized utilizing conjugation of biotin and streptavidin. Biotinylated NOVX protein or target molecules can be prepared from biotin-NHS (N-hydroxy-succinimide) using techniques well-known within the art (*e.g.*, biotinylation kit, Pierce Chemicals, Rockford, Ill.), and immobilized in the wells of streptavidin-coated 96 well plates (Pierce Chemical). Alternatively, antibodies reactive with NOVX protein or target molecules, but which do not interfere with binding of the NOVX protein to its target molecule, can be derivatized to the wells of the plate, and unbound target or NOVX protein trapped in the wells by antibody conjugation. Methods for detecting such complexes, in addition to those described above for the GST-immobilized complexes, include immunodetection of complexes using antibodies reactive with the NOVX protein or target molecule, as well as enzyme-linked assays that rely on detecting an enzymatic activity associated with the NOVX protein or target molecule.

In another embodiment, modulators of NOVX protein expression are identified in a method wherein a cell is contacted with a candidate compound and the expression of NOVX mRNA or protein in the cell is determined. The level of expression of NOVX mRNA or protein in the presence of the candidate compound is compared to the level of expression of NOVX mRNA or protein in the absence of the candidate compound. The candidate compound can then be identified as a modulator of NOVX mRNA or protein expression based upon this comparison. For example, when expression of NOVX mRNA or protein is greater (*i.e.*, statistically significantly greater) in the presence of the candidate compound than in its absence, the candidate compound is identified as a stimulator of NOVX mRNA or protein expression. Alternatively, when expression of NOVX mRNA or protein is less (statistically significantly less) in the presence of the candidate compound than in its absence, the candidate compound is identified as an inhibitor of NOVX mRNA or protein expression. The level of NOVX mRNA or protein expression in the cells can be determined by methods described herein for detecting NOVX mRNA or protein.

In yet another aspect of the invention, the NOVX proteins can be used as "bait proteins" in a two-hybrid assay or three hybrid assay (*see, e.g.*, U.S. Patent No. 5,283,317; Zervos, *et al.*, 1993. *Cell* 72: 223-232; Madura, *et al.*, 1993. *J. Biol. Chem.* 268: 12046-12054; Bartel, *et al.*, 1993. *Biotechniques* 14: 920-924; Iwabuchi, *et al.*, 1993. *Oncogene* 8: 1693-1696; and Brent WO 94/10300), to identify other proteins that bind to or interact with NOVX ("NOVX-binding proteins" or "NOVX-bp") and modulate NOVX activity. Such NOVX-binding proteins are also

likely to be involved in the propagation of signals by the NOVX proteins as, for example, upstream or downstream elements of the NOVX pathway.

The two-hybrid system is based on the modular nature of most transcription factors, which consist of separable DNA-binding and activation domains. Briefly, the assay utilizes two different DNA constructs. In one construct, the gene that codes for NOVX is fused to a gene encoding the DNA binding domain of a known transcription factor (*e.g.*, GAL-4). In the other construct, a DNA sequence, from a library of DNA sequences, that encodes an unidentified protein ("prey" or "sample") is fused to a gene that codes for the activation domain of the known transcription factor. If the "bait" and the "prey" proteins are able to interact, *in vivo*, forming an NOVX-dependent complex, the DNA-binding and activation domains of the transcription factor are brought into close proximity. This proximity allows transcription of a reporter gene (*e.g.*, LacZ) that is operably linked to a transcriptional regulatory site responsive to the transcription factor. Expression of the reporter gene can be detected and cell colonies containing the functional transcription factor can be isolated and used to obtain the cloned gene that encodes the protein which interacts with NOVX.

The invention further pertains to novel agents identified by the aforementioned screening assays and uses thereof for treatments as described herein.

Detection Assays

Portions or fragments of the cDNA sequences identified herein (and the corresponding complete gene sequences) can be used in numerous ways as polynucleotide reagents. By way of example, and not of limitation, these sequences can be used to: (*i*) map their respective genes on a chromosome; and, thus, locate gene regions associated with genetic disease; (*ii*) identify an individual from a minute biological sample (tissue typing); and (*iii*) aid in forensic identification of a biological sample. Some of these applications are described in the subsections, below.

Chromosome Mapping

Once the sequence (or a portion of the sequence) of a gene has been isolated, this sequence can be used to map the location of the gene on a chromosome. This process is called chromosome mapping. Accordingly, portions or fragments of the NOVX sequences, SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102, or fragments or derivatives thereof, can be used to map the location of the NOVX genes, respectively, on a chromosome. The mapping of the NOVX sequences to chromosomes is an important first step in correlating these sequences with genes associated with disease.

Briefly, NOVX genes can be mapped to chromosomes by preparing PCR primers (preferably 15-25 bp in length) from the NOVX sequences. Computer analysis of the NOVX, sequences can be used to rapidly select primers that do not span more than one exon in the genomic DNA, thus complicating the amplification process. These primers can then be used for PCR screening of somatic cell hybrids containing individual human chromosomes. Only those hybrids containing the human gene corresponding to the NOVX sequences will yield an amplified fragment.

Somatic cell hybrids are prepared by fusing somatic cells from different mammals (*e.g.*, human and mouse cells). As hybrids of human and mouse cells grow and divide, they gradually lose human chromosomes in random order, but retain the mouse chromosomes. By using media in which mouse cells cannot grow, because they lack a particular enzyme, but in which human cells can, the one human chromosome that contains the gene encoding the needed enzyme will be retained. By using various media, panels of hybrid cell lines can be established. Each cell line in a panel contains either a single human chromosome or a small number of human chromosomes, and a full set of mouse chromosomes, allowing easy mapping of individual genes to specific human chromosomes. *See, e.g.,* D'Eustachio, *et al.*, 1983. *Science* 220: 919-924. Somatic cell hybrids containing only fragments of human chromosomes can also be produced by using human chromosomes with translocations and deletions.

PCR mapping of somatic cell hybrids is a rapid procedure for assigning a particular sequence to a particular chromosome. Three or more sequences can be assigned per day using a single thermal cycler. Using the NOVX sequences to design oligonucleotide primers, sub-localization can be achieved with panels of fragments from specific chromosomes.

Fluorescence *in situ* hybridization (FISH) of a DNA sequence to a metaphase chromosomal spread can further be used to provide a precise chromosomal location in one step. Chromosome spreads can be made using cells whose division has been blocked in metaphase by a chemical like colcemid that disrupts the mitotic spindle. The chromosomes can be treated briefly with trypsin, and then stained with Giemsa. A pattern of light and dark bands develops on each chromosome, so that the chromosomes can be identified individually. The FISH technique can be used with a DNA sequence as short as 500 or 600 bases. However, clones larger than 1,000 bases have a higher likelihood of binding to a unique chromosomal location with sufficient signal intensity for simple detection. Preferably 1,000 bases, and more preferably 2,000 bases, will suffice to get good results at a reasonable amount of time. For a review of this technique, *see,* Verma, *et al.*, HUMAN CHROMOSOMES: A MANUAL OF BASIC TECHNIQUES (Pergamon Press, New York 1988).

Reagents for chromosome mapping can be used individually to mark a single chromosome or a single site on that chromosome, or panels of reagents can be used for marking multiple sites and/or multiple chromosomes. Reagents corresponding to noncoding regions of the genes actually are preferred for mapping purposes. Coding sequences are more likely to be conserved within gene families, thus increasing the chance of cross hybridizations during chromosomal mapping.

Once a sequence has been mapped to a precise chromosomal location, the physical position of the sequence on the chromosome can be correlated with genetic map data. Such data are found, *e.g.,* in McKusick, MENDELIAN INHERITANCE IN MAN, available on-line through Johns Hopkins University Welch Medical Library). The relationship between genes and disease, mapped to the same chromosomal region, can then be identified through linkage analysis (co-inheritance of physically adjacent genes), described in, *e.g.,* Egeland, *et al.*, 1987. *Nature*, 325: 783-787.

Moreover, differences in the DNA sequences between individuals affected and unaffected with a disease associated with the NOVX gene, can be determined. If a mutation is observed in some or all of the affected individuals but not in any unaffected individuals, then the mutation is likely to be the causative agent of the particular disease. Comparison of affected and unaffected individuals generally involves first looking for structural alterations in the chromosomes, such as deletions or translocations that are visible from chromosome spreads or detectable using PCR based on that DNA sequence. Ultimately, complete sequencing of genes from several individuals can be performed to confirm the presence of a mutation and to distinguish mutations from polymorphisms.

Tissue Typing

The NOVX sequences of the invention can also be used to identify individuals from minute biological samples. In this technique, an individual's genomic DNA is digested with one or more restriction enzymes, and probed on a Southern blot to yield unique bands for identification. The sequences of the invention are useful as additional DNA markers for RFLP ("restriction fragment length polymorphisms," described in U.S. Patent No. 5,272,057).

Furthermore, the sequences of the invention can be used to provide an alternative technique that determines the actual base-by-base DNA sequence of selected portions of an individual's genome. Thus, the NOVX sequences described herein can be used to prepare two PCR primers from the 5'- and 3'-termini of the sequences. These primers can then be used to amplify an individual's DNA and subsequently sequence it.

Panels of corresponding DNA sequences from individuals, prepared in this manner, can provide unique individual identifications, as each individual will have a unique set of such DNA sequences due to allelic differences. The sequences of the invention can be used to obtain such identification sequences from individuals and from tissue. The NOVX sequences of the invention uniquely represent portions of the human genome. Allelic variation occurs to some degree in the coding regions of these sequences, and to a greater degree in the noncoding regions. It is estimated that allelic variation between individual humans occurs with a frequency of about once per each 500 bases. Much of the allelic variation is due to single nucleotide polymorphisms (SNPs), which include restriction fragment length polymorphisms (RFLPs).

Each of the sequences described herein can, to some degree, be used as a standard against which DNA from an individual can be compared for identification purposes. Because greater numbers of polymorphisms occur in the noncoding regions, fewer sequences are necessary to differentiate individuals. The noncoding sequences can comfortably provide positive individual identification with a panel of perhaps 10 to 1,000 primers that each yield a noncoding amplified sequence of 100 bases. If predicted coding sequences, such as those in SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102 are used, a more appropriate number of primers for positive individual identification would be 500-2,000.

Predictive Medicine

The invention also pertains to the field of predictive medicine in which diagnostic assays, prognostic assays, pharmacogenomics, and monitoring clinical trials are used for prognostic (predictive) purposes to thereby treat an individual prophylactically. Accordingly, one aspect of the invention relates to diagnostic assays for determining NOVX protein and/or nucleic acid expression as well as NOVX activity, in the context of a biological sample (*e.g.*, blood, serum, cells, tissue) to thereby determine whether an individual is afflicted with a disease or disorder, or is at risk of developing a disorder, associated with aberrant NOVX expression or activity. The disorders include metabolic disorders, diabetes, obesity, infectious disease, anorexia, cancer-associated cachexia, cancer, neurodegenerative disorders, Alzheimer's Disease, Parkinson's Disorder, immune disorders, and hematopoietic disorders, and the various dyslipidemias, metabolic disturbances associated with obesity, the metabolic syndrome X and wasting disorders associated with chronic diseases and various cancers. The invention also provides for prognostic (or predictive) assays for determining whether an individual is at risk of developing a disorder associated with NOVX protein, nucleic acid expression or activity. For example, mutations in an NOVX gene can be assayed in a biological sample. Such assays can be used for prognostic or predictive purpose to thereby prophylactically treat an individual prior to the onset

of a disorder characterized by or associated with NOVX protein, nucleic acid expression, or biological activity.

Another aspect of the invention provides methods for determining NOVX protein, nucleic acid expression or activity in an individual to thereby select appropriate therapeutic or prophylactic agents for that individual (referred to herein as "pharmacogenomics"). Pharmacogenomics allows for the selection of agents (*e.g.*, drugs) for therapeutic or prophylactic treatment of an individual based on the genotype of the individual (*e.g.*, the genotype of the individual examined to determine the ability of the individual to respond to a particular agent.)

Yet another aspect of the invention pertains to monitoring the influence of agents (*e.g.*, drugs, compounds) on the expression or activity of NOVX in clinical trials.

These and other agents are described in further detail in the following sections.

Diagnostic Assays

An exemplary method for detecting the presence or absence of NOVX in a biological sample involves obtaining a biological sample from a test subject and contacting the biological sample with a compound or an agent capable of detecting NOVX protein or nucleic acid (*e.g.*, mRNA, genomic DNA) that encodes NOVX protein such that the presence of NOVX is detected in the biological sample. An agent for detecting NOVX mRNA or genomic DNA is a labeled nucleic acid probe capable of hybridizing to NOVX mRNA or genomic DNA. The nucleic acid probe can be, for example, a full-length NOVX nucleic acid, such as the nucleic acid of SEQ ID NOS:2, 9, 11, 19, 27, 35, 43, 51, 53, 61, 63, 65, 71, 73, 75, 83, 90, 92, 100 and 102, or a portion thereof, such as an oligonucleotide of at least 15, 30, 50, 100, 250 or 500 nucleotides in length and sufficient to specifically hybridize under stringent conditions to NOVX mRNA or genomic DNA. Other suitable probes for use in the diagnostic assays of the invention are described herein.

An agent for detecting NOVX protein is an antibody capable of binding to NOVX protein, preferably an antibody with a detectable label. Antibodies can be polyclonal, or more preferably, monoclonal. An intact antibody, or a fragment thereof (*e.g.*, Fab or $F(ab')_2$) can be used. The term "labeled", with regard to the probe or antibody, is intended to encompass direct labeling of the probe or antibody by coupling (*i.e.*, physically linking) a detectable substance to the probe or antibody, as well as indirect labeling of the probe or antibody by reactivity with another reagent that is directly labeled. Examples of indirect labeling include detection of a primary antibody using a fluorescently-labeled secondary antibody and end-labeling of a DNA probe with biotin such that it can be detected with fluorescently-labeled streptavidin. The term "biological sample" is intended to include tissues, cells and biological fluids isolated from a

subject, as well as tissues, cells and fluids present within a subject. That is, the detection method of the invention can be used to detect NOVX mRNA, protein, or genomic DNA in a biological sample *in vitro* as well as *in vivo*. For example, *in vitro* techniques for detection of NOVX mRNA include Northern hybridizations and *in situ* hybridizations. *In vitro* techniques for detection of NOVX protein include enzyme linked immunosorbent assays (ELISAs), Western blots, immunoprecipitations, and immunofluorescence. *In vitro* techniques for detection of NOVX genomic DNA include Southern hybridizations. Furthermore. *in vivo* techniques for detection of NOVX protein include introducing into a subject a labeled anti-NOVX antibody. For example, the antibody can be labeled with a radioactive marker whose presence and location in a subject can be detected by standard imaging techniques.

In one embodiment, the biological sample contains protein molecules from the test subject. Alternatively, the biological sample can contain mRNA molecules from the test subject or genomic DNA molecules from the test subject. A preferred biological sample is a peripheral blood leukocyte sample isolated by conventional means from a subject.

In another embodiment, the methods further involve obtaining a control biological sample from a control subject, contacting the control sample with a compound or agent capable of detecting NOVX protein, mRNA, or genomic DNA, such that the presence of NOVX protein, mRNA or genomic DNA is detected in the biological sample, and comparing the presence of NOVX protein, mRNA or genomic DNA in the control sample with the presence of NOVX protein, mRNA or genomic DNA in the test sample.

The invention also encompasses kits for detecting the presence of NOVX in a biological sample. For example, the kit can comprise: a labeled compound or agent capable of detecting NOVX protein or mRNA in a biological sample; means for determining the amount of NOVX in the sample; and means for comparing the amount of NOVX in the sample with a standard. The compound or agent can be packaged in a suitable container. The kit can further comprise instructions for using the kit to detect NOVX protein or nucleic acid.

Prognostic Assays

The diagnostic methods described herein can furthermore be utilized to identify subjects having or at risk of developing a disease or disorder associated with aberrant NOVX expression or activity. For example, the assays described herein, such as the preceding diagnostic assays or the following assays, can be utilized to identify a subject having or at risk of developing a disorder associated with NOVX protein, nucleic acid expression or activity. Alternatively, the prognostic assays can be utilized to identify a subject having or at risk for developing a disease or disorder. Thus, the invention provides a method for identifying a disease or disorder

associated with aberrant NOVX expression or activity in which a test sample is obtained from a subject and NOVX protein or nucleic acid (*e.g.*, mRNA, genomic DNA) is detected, wherein the presence of NOVX protein or nucleic acid is diagnostic for a subject having or at risk of developing a disease or disorder associated with aberrant NOVX expression or activity. As used herein, a "test sample" refers to a biological sample obtained from a subject of interest. For example, a test sample can be a biological fluid (*e.g.* serum), cell sample, or tissue.

Furthermore, the prognostic assays described herein can be used to determine whether a subject can be administered an agent (*e.g.*, an agonist, antagonist, peptidomimetic, protein, peptide, nucleic acid, small molecule, or other drug candidate) to treat a disease or disorder associated with aberrant NOVX expression or activity. For example, such methods can be used to determine whether a subject can be effectively treated with an agent for a disorder. Thus, the invention provides methods for determining whether a subject can be effectively treated with an agent for a disorder associated with aberrant NOVX expression or activity in which a test sample is obtained and NOVX protein or nucleic acid is detected (*e.g.*, wherein the presence of NOVX protein or nucleic acid is diagnostic for a subject that can be administered the agent to treat a disorder associated with aberrant NOVX expression or activity).

The methods of the invention can also be used to detect genetic lesions in an NOVX gene, thereby determining if a subject with the lesioned gene is at risk for a disorder characterized by aberrant cell proliferation and/or differentiation. In various embodiments, the methods include detecting, in a sample of cells from the subject, the presence or absence of a genetic lesion characterized by at least one of an alteration affecting the integrity of a gene encoding an NOVX-protein, or the misexpression of the NOVX gene. For example, such genetic lesions can be detected by ascertaining the existence of at least one of: (*i*) a deletion of one or more nucleotides from an NOVX gene; (*ii*) an addition of one or more nucleotides to an NOVX gene; (*iii*) a substitution of one or more nucleotides of an NOVX gene, (*iv*) a chromosomal rearrangement of an NOVX gene; (*v*) an alteration in the level of a messenger RNA transcript of an NOVX gene, (*vi*) aberrant modification of an NOVX gene, such as of the methylation pattern of the genomic DNA, (*vii*) the presence of a non-wild-type splicing pattern of a messenger RNA transcript of an NOVX gene, (*viii*) a non-wild-type level of an NOVX protein, (*ix*) allelic loss of an NOVX gene, and (*x*) inappropriate post-translational modification of an NOVX protein. As described herein, there are a large number of assay techniques known in the art which can be used for detecting lesions in an NOVX gene. A preferred biological sample is a peripheral blood leukocyte sample isolated by conventional means from a subject. However, any biological sample containing nucleated cells may be used, including, for example, buccal mucosal cells.

In certain embodiments, detection of the lesion involves the use of a probe/primer in a polymerase chain reaction (PCR) (*see, e.g.*, U.S. Patent Nos. 4,683,195 and 4,683,202), such as anchor PCR or RACE PCR, or, alternatively, in a ligation chain reaction (LCR) (*see, e.g.*, Landegran, *et al.*, 1988. *Science* 241: 1077-1080; and Nakazawa, *et al.*, 1994. *Proc. Natl. Acad. Sci. USA* 91: 360-364), the latter of which can be particularly useful for detecting point mutations in the NOVX-gene (*see,* Abravaya, *et al.*, 1995. *Nucl. Acids Res.* 23: 675-682). This method can include the steps of collecting a sample of cells from a patient, isolating nucleic acid (*e.g.*, genomic, mRNA or both) from the cells of the sample, contacting the nucleic acid sample with one or more primers that specifically hybridize to an NOVX gene under conditions such that hybridization and amplification of the NOVX gene (if present) occurs, and detecting the presence or absence of an amplification product, or detecting the size of the amplification product and comparing the length to a control sample. It is anticipated that PCR and/or LCR may be desirable to use as a preliminary amplification step in conjunction with any of the techniques used for detecting mutations described herein.

Alternative amplification methods include: self sustained sequence replication (*see,* Guatelli, *et al.*, 1990. *Proc. Natl. Acad. Sci. USA* 87: 1874-1878), transcriptional amplification system (*see,* Kwoh, *et al.*, 1989. *Proc. Natl. Acad. Sci. USA* 86: 1173-1177); Qβ Replicase (*see,* Lizardi, *et al*, 1988. *BioTechnology* 6: 1197), or any other nucleic acid amplification method, followed by the detection of the amplified molecules using techniques well known to those of skill in the art. These detection schemes are especially useful for the detection of nucleic acid molecules if such molecules are present in very low numbers.

In an alternative embodiment, mutations in an NOVX gene from a sample cell can be identified by alterations in restriction enzyme cleavage patterns. For example, sample and control DNA is isolated, amplified (optionally), digested with one or more restriction endonucleases, and fragment length sizes are determined by gel electrophoresis and compared. Differences in fragment length sizes between sample and control DNA indicates mutations in the sample DNA. Moreover, the use of sequence specific ribozymes (*see, e.g.,* U.S. Patent No. 5,493,531) can be used to score for the presence of specific mutations by development or loss of a ribozyme cleavage site.

In other embodiments, genetic mutations in NOVX can be identified by hybridizing a sample and control nucleic acids, *e.g.*, DNA or RNA, to high-density arrays containing hundreds or thousands of oligonucleotides probes. *See, e.g.,* Cronin, *et al.*, 1996. *Human Mutation* 7: 244-255; Kozal, *et al.*, 1996. *Nat. Med.* 2: 753-759. For example, genetic mutations in NOVX can be identified in two dimensional arrays containing light-generated DNA probes as described in Cronin, *et al.*, *supra.* Briefly, a first hybridization array of probes can be used to scan through

long stretches of DNA in a sample and control to identify base changes between the sequences by making linear arrays of sequential overlapping probes. This step allows the identification of point mutations. This is followed by a second hybridization array that allows the characterization of specific mutations by using smaller, specialized probe arrays complementary to all variants or mutations detected. Each mutation array is composed of parallel probe sets, one complementary to the wild-type gene and the other complementary to the mutant gene.

In yet another embodiment, any of a variety of sequencing reactions known in the art can be used to directly sequence the NOVX gene and detect mutations by comparing the sequence of the sample NOVX with the corresponding wild-type (control) sequence. Examples of sequencing reactions include those based on techniques developed by Maxim and Gilbert, 1977. *Proc. Natl. Acad. Sci USA* 74: 560 or Sanger, 1977. *Proc. Natl. Acad. Sci. USA* 74: 5463. It is also contemplated that any of a variety of automated sequencing procedures can be utilized when performing the diagnostic assays (*see. e.g.,* Naeve, *et al.*, 1995. *Biotechniques* 19: 448), including sequencing by mass spectrometry (see, *e.g.*, PCT International Publication No. WO 94/16101; Cohen, *et al.*, 1996. *Adv. Chromatography* 36: 127-162; and Griffin, *et al.*, 1993. *Appl. Biochem. Biotechnol.* 38: 147-159).

Other methods for detecting mutations in the NOVX gene include methods in which protection from cleavage agents is used to detect mismatched bases in RNA/RNA or RNA/DNA heteroduplexes. *See, e.g.,* Myers, *et al.*, 1985. *Science* 230: 1242. In general, the art technique of "mismatch cleavage" starts by providing heteroduplexes of formed by hybridizing (labeled) RNA or DNA containing the wild-type NOVX sequence with potentially mutant RNA or DNA obtained from a tissue sample. The double-stranded duplexes are treated with an agent that cleaves single-stranded regions of the duplex such as which will exist due to basepair mismatches between the control and sample strands. For instance, RNA/DNA duplexes can be treated with RNase and DNA/DNA hybrids treated with S_1 nuclease to enzymatically digesting the mismatched regions. In other embodiments, either DNA/DNA or RNA/DNA duplexes can be treated with hydroxylamine or osmium tetroxide and with piperidine in order to digest mismatched regions. After digestion of the mismatched regions, the resulting material is then separated by size on denaturing polyacrylamide gels to determine the site of mutation. *See, e.g.,* Cotton, *et al.,* 1988. *Proc. Natl. Acad. Sci. USA* 85: 4397; Saleeba, *et al.,* 1992. *Methods Enzymol.* 217: 286-295. In an embodiment, the control DNA or RNA can be labeled for detection.

In still another embodiment, the mismatch cleavage reaction employs one or more proteins that recognize mismatched base pairs in double-stranded DNA (so called "DNA mismatch repair" enzymes) in defined systems for detecting and mapping point mutations in

NOVX cDNAs obtained from samples of cells. For example, the mutY enzyme of *E. coli* cleaves A at G/A mismatches and the thymidine DNA glycosylase from HeLa cells cleaves T at G/T mismatches. *See, e.g.,* Hsu, *et al.*, 1994. *Carcinogenesis* 15: 1657-1662. According to an exemplary embodiment, a probe based on an NOVX sequence, *e.g.*, a wild-type NOVX sequence, is hybridized to a cDNA or other DNA product from a test cell(s). The duplex is treated with a DNA mismatch repair enzyme, and the cleavage products, if any, can be detected from electrophoresis protocols or the like. *See, e.g.,* U.S. Patent No. 5,459,039.

In other embodiments, alterations in electrophoretic mobility will be used to identify mutations in NOVX genes. For example, single strand conformation polymorphism (SSCP) may be used to detect differences in electrophoretic mobility between mutant and wild type nucleic acids. *See, e.g.,* Orita, *et al.*, 1989. *Proc. Natl. Acad. Sci. USA*: 86: 2766; Cotton, 1993. *Mutat. Res.* 285: 125-144; Hayashi, 1992. *Genet. Anal. Tech. Appl.* 9: 73-79. Single-stranded DNA fragments of sample and control NOVX nucleic acids will be denatured and allowed to renature. The secondary structure of single-stranded nucleic acids varies according to sequence, the resulting alteration in electrophoretic mobility enables the detection of even a single base change. The DNA fragments may be labeled or detected with labeled probes. The sensitivity of the assay may be enhanced by using RNA (rather than DNA), in which the secondary structure is more sensitive to a change in sequence. In one embodiment, the subject method utilizes heteroduplex analysis to separate double stranded heteroduplex molecules on the basis of changes in electrophoretic mobility. *See, e.g.,* Keen, *et al.*, 1991. *Trends Genet.* 7: 5.

In yet another embodiment, the movement of mutant or wild-type fragments in polyacrylamide gels containing a gradient of denaturant is assayed using denaturing gradient gel electrophoresis (DGGE). *See, e.g.,* Myers, *et al.,* 1985. *Nature* 313: 495. When DGGE is used as the method of analysis, DNA will be modified to insure that it does not completely denature, for example by adding a GC clamp of approximately 40 bp of high-melting GC-rich DNA by PCR. In a further embodiment, a temperature gradient is used in place of a denaturing gradient to identify differences in the mobility of control and sample DNA. *See, e.g.,* Rosenbaum and Reissner, 1987. *Biophys. Chem.* 265: 12753.

Examples of other techniques for detecting point mutations include, but are not limited to, selective oligonucleotide hybridization, selective amplification, or selective primer extension. For example, oligonucleotide primers may be prepared in which the known mutation is placed centrally and then hybridized to target DNA under conditions that permit hybridization only if a perfect match is found. *See, e.g.,* Saiki, *et al.*, 1986. *Nature* 324: 163; Saiki, *et al.*, 1989. *Proc. Natl. Acad. Sci. USA* 86: 6230. Such allele specific oligonucleotides are hybridized to PCR

amplified target DNA or a number of different mutations when the oligonucleotides are attached to the hybridizing membrane and hybridized with labeled target DNA.

Alternatively, allele specific amplification technology that depends on selective PCR amplification may be used in conjunction with the instant invention. Oligonucleotides used as primers for specific amplification may carry the mutation of interest in the center of the molecule (so that amplification depends on differential hybridization; *see, e.g.*, Gibbs. *et al.*, 1989. *Nucl. Acids Res.* 17: 2437-2448) or at the extreme 3'-terminus of one primer where, under appropriate conditions, mismatch can prevent, or reduce polymerase extension (*see, e.g.,* Prossner, 1993. *Tibtech.* 11: 238). In addition it may be desirable to introduce a novel restriction site in the region of the mutation to create cleavage-based detection. *See, e.g.,* Gasparini, *et al.*, 1992. *Mol. Cell Probes* 6: 1. It is anticipated that in certain embodiments amplification may also be performed using *Taq* ligase for amplification. *See, e.g.,* Barany, 1991. *Proc. Natl. Acad. Sci. USA* 88: 189. In such cases, ligation will occur only if there is a perfect match at the 3'-terminus of the 5' sequence, making it possible to detect the presence of a known mutation at a specific site by looking for the presence or absence of amplification.

The methods described herein may be performed, for example, by utilizing pre-packaged diagnostic kits comprising at least one probe nucleic acid or antibody reagent described herein, which may be conveniently used, *e.g.*, in clinical settings to diagnose patients exhibiting symptoms or family history of a disease or illness involving an NOVX gene.

Furthermore, any cell type or tissue, preferably peripheral blood leukocytes, in which NOVX is expressed may be utilized in the prognostic assays described herein. However, any biological sample containing nucleated cells may be used, including, for example, buccal mucosal cells.

Pharmacogenomics

Agents, or modulators that have a stimulatory or inhibitory effect on NOVX activity (*e.g.*, NOVX gene expression), as identified by a screening assay described herein can be administered to individuals to treat (prophylactically or therapeutically) disorders (The disorders include metabolic disorders, diabetes, obesity, infectious disease, anorexia, cancer-associated cachexia, cancer, neurodegenerative disorders, Alzheimer's Disease, Parkinson's Disorder, immune disorders, and hematopoietic disorders, and the various dyslipidemias, metabolic disturbances associated with obesity, the metabolic syndrome X and wasting disorders associated with chronic diseases and various cancers.) In conjunction with such treatment, the pharmacogenomics (*i.e.*, the study of the relationship between an individual's genotype and that individual's response to a foreign compound or drug) of the individual may be considered.

Differences in metabolism of therapeutics can lead to severe toxicity or therapeutic failure by altering the relation between dose and blood concentration of the pharmacologically active drug. Thus, the pharmacogenomics of the individual permits the selection of effective agents (*e.g.*, drugs) for prophylactic or therapeutic treatments based on a consideration of the individual's genotype. Such pharmacogenomics can further be used to determine appropriate dosages and therapeutic regimens. Accordingly, the activity of NOVX protein, expression of NOVX nucleic acid, or mutation content of NOVX genes in an individual can be determined to thereby select appropriate agent(s) for therapeutic or prophylactic treatment of the individual.

Pharmacogenomics deals with clinically significant hereditary variations in the response to drugs due to altered drug disposition and abnormal action in affected persons. See *e.g.*, Eichelbaum, 1996. *Clin. Exp. Pharmacol. Physiol.*, 23: 983-985; Linder, 1997. *Clin. Chem.*, 43: 254-266. In general, two types of pharmacogenetic conditions can be differentiated. Genetic conditions transmitted as a single factor altering the way drugs act on the body (altered drug action) or genetic conditions transmitted as single factors altering the way the body acts on drugs (altered drug metabolism). These pharmacogenetic conditions can occur either as rare defects or as polymorphisms. For example, glucose-6-phosphate dehydrogenase (G6PD) deficiency is a common inherited enzymopathy in which the main clinical complication is hemolysis after ingestion of oxidant drugs (anti-malarials, sulfonamides, analgesics, nitrofurans) and consumption of fava beans.

As an illustrative embodiment, the activity of drug metabolizing enzymes is a major determinant of both the intensity and duration of drug action. The discovery of genetic polymorphisms of drug metabolizing enzymes (*e.g.*, N-acetyltransferase 2 (NAT 2) and cytochrome P450 enzymes CYP2D6 and CYP2C19) has provided an explanation as to why some patients do not obtain the expected drug effects or show exaggerated drug response and serious toxicity after taking the standard and safe dose of a drug. These polymorphisms are expressed in two phenotypes in the population, the extensive metabolizer (EM) and poor metabolizer (PM). The prevalence of PM is different among different populations. For example, the gene coding for CYP2D6 is highly polymorphic and several mutations have been identified in PM, which all lead to the absence of functional CYP2D6. Poor metabolizers of CYP2D6 and CYP2C19 quite frequently experience exaggerated drug response and side effects when they receive standard doses. If a metabolite is the active therapeutic moiety, PM show no therapeutic response, as demonstrated for the analgesic effect of codeine mediated by its CYP2D6-formed metabolite morphine. At the other extreme are the so called ultra-rapid metabolizers who do not respond to standard doses. Recently, the molecular basis of ultra-rapid metabolism has been identified to be due to CYP2D6 gene amplification.

Thus, the activity of NOVX protein, expression of NOVX nucleic acid, or mutation content of NOVX genes in an individual can be determined to thereby select appropriate agent(s) for therapeutic or prophylactic treatment of the individual. In addition, pharmacogenetic studies can be used to apply genotyping of polymorphic alleles encoding drug-metabolizing enzymes to the identification of an individual's drug responsiveness phenotype. This knowledge, when applied to dosing or drug selection, can avoid adverse reactions or therapeutic failure and thus enhance therapeutic or prophylactic efficiency when treating a subject with an NOVX modulator, such as a modulator identified by one of the exemplary screening assays described herein.

Monitoring of Effects During Clinical Trials

Monitoring the influence of agents (*e.g.*, drugs, compounds) on the expression or activity of NOVX (*e.g.*, the ability to modulate aberrant cell proliferation and/or differentiation) can be applied not only in basic drug screening, but also in clinical trials. For example, the effectiveness of an agent determined by a screening assay as described herein to increase NOVX gene expression, protein levels, or upregulate NOVX activity, can be monitored in clinical trails of subjects exhibiting decreased NOVX gene expression, protein levels, or downregulated NOVX activity. Alternatively, the effectiveness of an agent determined by a screening assay to decrease NOVX gene expression, protein levels, or downregulate NOVX activity, can be monitored in clinical trails of subjects exhibiting increased NOVX gene expression, protein levels, or upregulated NOVX activity. In such clinical trials, the expression or activity of NOVX and, preferably, other genes that have been implicated in, for example, a cellular proliferation or immune disorder can be used as a "read out" or markers of the immune responsiveness of a particular cell.

By way of example, and not of limitation, genes, including NOVX, that are modulated in cells by treatment with an agent (*e.g.*, compound, drug or small molecule) that modulates NOVX activity (*e.g.*, identified in a screening assay as described herein) can be identified. Thus, to study the effect of agents on cellular proliferation disorders, for example, in a clinical trial, cells can be isolated and RNA prepared and analyzed for the levels of expression of NOVX and other genes implicated in the disorder. The levels of gene expression (*i.e.*, a gene expression pattern) can be quantified by Northern blot analysis or RT-PCR, as described herein, or alternatively by measuring the amount of protein produced, by one of the methods as described herein, or by measuring the levels of activity of NOVX or other genes. In this manner, the gene expression pattern can serve as a marker, indicative of the physiological response of the cells to the agent.

Accordingly, this response state may be determined before, and at various points during, treatment of the individual with the agent.

In one embodiment, the invention provides a method for monitoring the effectiveness of treatment of a subject with an agent (*e.g.*, an agonist, antagonist, protein, peptide, peptidomimetic, nucleic acid, small molecule, or other drug candidate identified by the screening assays described herein) comprising the steps of (*i*) obtaining a pre-administration sample from a subject prior to administration of the agent; (*ii*) detecting the level of expression of an NOVX protein, mRNA, or genomic DNA in the preadministration sample; (*iii*) obtaining one or more post-administration samples from the subject; (*iv*) detecting the level of expression or activity of the NOVX protein, mRNA, or genomic DNA in the post-administration samples; (*v*) comparing the level of expression or activity of the NOVX protein, mRNA, or genomic DNA in the pre-administration sample with the NOVX protein, mRNA, or genomic DNA in the post administration sample or samples; and (*vi*) altering the administration of the agent to the subject accordingly. For example, increased administration of the agent may be desirable to increase the expression or activity of NOVX to higher levels than detected, *i.e.*, to increase the effectiveness of the agent. Alternatively, decreased administration of the agent may be desirable to decrease expression or activity of NOVX to lower levels than detected, *i.e* , to decrease the effectiveness of the agent.

Methods of Treatment

The invention provides for both prophylactic and therapeutic methods of treating a subject at risk of (or susceptible to) a disorder or having a disorder associated with aberrant NOVX expression or activity. The disorders include cardiomyopathy, atherosclerosis, hypertension, congenital heart defects, aortic stenosis, atrial septal defect (ASD), atrioventricular (A-V) canal defect, ductus arteriosus, pulmonary stenosis, subaortic stenosis, ventricular septal defect (VSD), valve diseases, tuberous sclerosis, scleroderma, obesity, transplantation, adrenoleukodystrophy, congenital adrenal hyperplasia, prostate cancer, neoplasm; adenocarcinoma, lymphoma, uterus cancer, fertility, hemophilia, hypercoagulation, idiopathic thrombocytopenic purpura, immunodeficiencies, graft versus host disease, AIDS, bronchial asthma, Crohn's disease; multiple sclerosis, treatment of Albright Hereditary Ostoeodystrophy, and other diseases, disorders and conditions of the like.

These methods of treatment will be discussed more fully, below.

Disease and Disorders

Diseases and disorders that are characterized by increased (relative to a subject not suffering from the disease or disorder) levels or biological activity may be treated with

Therapeutics that antagonize (*i.e.*, reduce or inhibit) activity. Therapeutics that antagonize activity may be administered in a therapeutic or prophylactic manner. Therapeutics that may be utilized include, but are not limited to: (*i*) an aforementioned peptide, or analogs, derivatives, fragments or homologs thereof; (*ii*) antibodies to an aforementioned peptide; (*iii*) nucleic acids encoding an aforementioned peptide; (*iv*) administration of antisense nucleic acid and nucleic acids that are "dysfunctional" (*i.e.*, due to a heterologous insertion within the coding sequences of coding sequences to an aforementioned peptide) that are utilized to "knockout" endogenous function of an aforementioned peptide by homologous recombination (*see, e.g.*, Capecchi, 1989. *Science* 244: 1288-1292); or (*v*) modulators (*i.e* , inhibitors, agonists and antagonists, including additional peptide mimetic of the invention or antibodies specific to a peptide of the invention) that alter the interaction between an aforementioned peptide and its binding partner.

Diseases and disorders that are characterized by decreased (relative to a subject not suffering from the disease or disorder) levels or biological activity may be treated with Therapeutics that increase (*i.e.*, are agonists to) activity. Therapeutics that upregulate activity may be administered in a therapeutic or prophylactic manner. Therapeutics that may be utilized include, but are not limited to, an aforementioned peptide, or analogs, derivatives, fragments or homologs thereof; or an agonist that increases bioavailability.

Increased or decreased levels can be readily detected by quantifying peptide and/or RNA, by obtaining a patient tissue sample (*e.g.*, from biopsy tissue) and assaying it *in vitro* for RNA or peptide levels, structure and/or activity of the expressed peptides (or mRNAs of an aforementioned peptide). Methods that are well-known within the art include, but are not limited to, immunoassays (*e.g.*, by Western blot analysis, immunoprecipitation followed by sodium dodecyl sulfate (SDS) polyacrylamide gel electrophoresis, immunocytochemistry, etc.) and/or hybridization assays to detect expression of mRNAs (*e.g.*, Northern assays, dot blots, *in situ* hybridization, and the like).

Prophylactic Methods

In one aspect, the invention provides a method for preventing, in a subject, a disease or condition associated with an aberrant NOVX expression or activity, by administering to the subject an agent that modulates NOVX expression or at least one NOVX activity. Subjects at risk for a disease that is caused or contributed to by aberrant NOVX expression or activity can be identified by, for example, any or a combination of diagnostic or prognostic assays as described herein. Administration of a prophylactic agent can occur prior to the manifestation of symptoms characteristic of the NOVX aberrancy, such that a disease or disorder is prevented or, alternatively, delayed in its progression. Depending upon the type of NOVX aberrancy, for example, an NOVX agonist or NOVX antagonist agent can be used for treating the subject. The

appropriate agent can be determined based on screening assays described herein. The prophylactic methods of the invention are further discussed in the following subsections.

Therapeutic Methods

Another aspect of the invention pertains to methods of modulating NOVX expression or activity for therapeutic purposes. The modulatory method of the invention involves contacting a cell with an agent that modulates one or more of the activities of NOVX protein activity associated with the cell. An agent that modulates NOVX protein activity can be an agent as described herein, such as a nucleic acid or a protein, a naturally-occurring cognate ligand of an NOVX protein, a peptide, an NOVX peptidomimetic, or other small molecule. In one embodiment, the agent stimulates one or more NOVX protein activity. Examples of such stimulatory agents include active NOVX protein and a nucleic acid molecule encoding NOVX that has been introduced into the cell. In another embodiment, the agent inhibits one or more NOVX protein activity. Examples of such inhibitory agents include antisense NOVX nucleic acid molecules and anti-NOVX antibodies. These modulatory methods can be performed *in vitro* (*e.g.*, by culturing the cell with the agent) or, alternatively, *in vivo* (*e.g.*, by administering the agent to a subject). As such, the invention provides methods of treating an individual afflicted with a disease or disorder characterized by aberrant expression or activity of an NOVX protein or nucleic acid molecule. In one embodiment, the method involves administering an agent (*e.g.*, an agent identified by a screening assay described herein), or combination of agents that modulates (*e.g.*, up-regulates or down-regulates) NOVX expression or activity. In another embodiment, the method involves administering an NOVX protein or nucleic acid molecule as therapy to compensate for reduced or aberrant NOVX expression or activity.

Stimulation of NOVX activity is desirable *in situ*ations in which NOVX is abnormally downregulated and/or in which increased NOVX activity is likely to have a beneficial effect. One example of such a situation is where a subject has a disorder characterized by aberrant cell proliferation and/or differentiation (*e.g.*, cancer or immune associated disorders). Another example of such a situation is where the subject has a gestational disease (*e.g.*, preclampsia).

Determination of the Biological Effect of the Therapeutic

In various embodiments of the invention, suitable *in vitro* or *in vivo* assays are performed to determine the effect of a specific Therapeutic and whether its administration is indicated for treatment of the affected tissue.

In various specific embodiments, *in vitro* assays may be performed with representative cells of the type(s) involved in the patient's disorder, to determine if a given Therapeutic exerts the desired effect upon the cell type(s). Compounds for use in therapy may be tested in suitable

animal model systems including, but not limited to rats, mice, chicken, cows, monkeys, rabbits, and the like, prior to testing in human subjects. Similarly, for *in vivo* testing, any of the animal model system known in the art may be used prior to administration to human subjects.

Prophylactic and Therapeutic Uses of the Compositions of the Invention

The NOVX nucleic acids and proteins of the invention are useful in potential prophylactic and therapeutic applications implicated in a variety of disorders including, but not limited to: metabolic disorders, diabetes, obesity, infectious disease, anorexia, cancer-associated cancer, neurodegenerative disorders, Alzheimer's Disease, Parkinson's Disorder, immune disorders, hematopoietic disorders, and the various dyslipidemias, metabolic disturbances associated with obesity, the metabolic syndrome X and wasting disorders associated with chronic diseases and various cancers.

As an example, a cDNA encoding the NOVX protein of the invention may be useful in gene therapy, and the protein may be useful when administered to a subject in need thereof. By way of non-limiting example, the compositions of the invention will have efficacy for treatment of patients suffering from: metabolic disorders, diabetes, obesity, infectious disease, anorexia, cancer-associated cachexia, cancer, neurodegenerative disorders, Alzheimer's Disease, Parkinson's Disorder, immune disorders, hematopoietic disorders, and the various dyslipidemias.

Both the novel nucleic acid encoding the NOVX protein, and the NOVX protein of the invention, or fragments thereof, may also be useful in diagnostic applications, wherein the presence or amount of the nucleic acid or the protein are to be assessed. A further use could be as an anti-bacterial molecule (*i.e.,* some peptides have been found to possess anti-bacterial properties). These materials are further useful in the generation of antibodies, which immunospecifically-bind to the novel substances of the invention for use in therapeutic or diagnostic methods.

The invention will be further described in the following examples, which do not limit the scope of the invention described in the claims.

EXAMPLES

Example 1. Identification of NOVX clones

The novel NOVX target sequences identified in the present invention were subjected to the exon linking process to confirm the sequence. PCR primers were designed by starting at the most upstream sequence available, for the forward primer, and at the most downstream sequence available for the reverse primer. Table 17A shows the sequences of the PCR primers used for obtaining different clones. In each case, the sequence was examined, walking inward from the

respective termini toward the coding sequence, until a suitable sequence that is either unique or highly selective was encountered, or, in the case of the reverse primer, until the stop codon was reached. Such primers were designed based on in silico predictions for the full length cDNA, part (one or more exons) of the DNA or protein sequence of the target sequence, or by translated homology of the predicted exons to closely related human sequences from other species. These primers were then employed in PCR amplification based on the following pool of human cDNAs: adrenal gland, bone marrow, brain - amygdala, brain - cerebellum, brain - hippocampus, brain - substantia nigra, brain - thalamus, brain -whole, fetal brain, fetal kidney, fetal liver, fetal lung, heart, kidney, lymphoma - Raji, mammary gland, pancreas, pituitary gland, placenta, prostate, salivary gland, skeletal muscle, small intestine, spinal cord, spleen, stomach, testis, thyroid, trachea, uterus. Usually the resulting amplicons were gel purified, cloned and sequenced to high redundancy. The PCR product derived from exon linking was cloned into the pCR2.1 vector from Invitrogen. The resulting bacterial clone has an insert covering the entire open reading frame cloned into the pCR2.1 vector. Table 17B shows a list of these bacterial clones. The resulting sequences from all clones were assembled with themselves, with other fragments in CuraGen Corporation's database and with public ESTs. Fragments and ESTs were included as components for an assembly when the extent of their identity with another component of the assembly was at least 95% over 50 bp. In addition, sequence traces were evaluated manually and edited for corrections if appropriate. These procedures provide the sequence reported herein.

Table 17A. PCR Primers for Exon Linking

NOVX Clone	Primer 1 (5' - 3')	SEQ ID NO	Primer 2 (5' - 3')	SEQ ID NO
NOV2	TGGCTTGATGATATGTGCCTGTAG	108	TTATAGTACGAGCAAGAACTTTGG	109
NOV3	TTATTGACAGTTTATCCTGCCGCACCT	110	AACTACTCGTGAGGCTGAGGCAGGAG	111
NOV4-1	CAATCCTTGCGTGTCCTTGCAGTC	112	AGCAAGCAAAATCAGGATGTTTTCCTC	113
NOV4-2	CAATCCTTGCGTGTCCTTGCAGTC	114	AGCAAGCAAAATCAGGATGTTTTCCTC	115
NOV10b	GCTACCTTCACCACCTCCTGCTGT	116	AAGTGCAGACCTATAGGCCAATACAGG	117
NOV10c	AGAACCCAAGGCTCCCTGGATT	118	CATGGAATTATTCAAATTTGCTCTG	119
NOV15	GTAGCCACAAGACCGGGTCCG	120	CCCTGGCCTCTTGGAACTGCTTGAT	121
NOV16	CCGCTGGCCGAGAGGCTGA	122	TGTTTAAAGCATTAATAAA	123

Physical clone: Exons were predicted by homology and the intron/exon boundaries were determined using standard genetic rules. Exons were further selected and refined by means of similarity determination using multiple BLAST (for example, tBlastN, BlastX, and BlastN) searches, and, in some instances, GeneScan and Grail. Expressed sequences from both public and proprietary databases were also added when available to further define and complete the gene sequence. The DNA sequence was then manually corrected for apparent inconsistencies thereby obtaining the sequences encoding the full-length protein.

Table 17B. Physical Clones for PCR products

NOVX Clone	Bacterial Clone	
NOV2	Physical clone:	110021::COR24CS059.698230.G1
NOV3	Physical clone:	104046::COR24SC113.698230.C13
NOV4	Physical clone:	110189::COR24SC128.698230.M23
NOV10b	Physical clone:	112812::COR100340173.698230.J3
NOV10c	Physical clone:	128970::80083680.698655.M23
NOV12	Physical clone:	112818::COR87917235.698230.N1
NOV15	Physical clone:	112824::COR100399281.698230.B6
NOV16	Physical clone:	112826::COR101330077.698230.F18.

Example 2. Quantitative expression analysis of clones in various cells and tissues

The quantitative expression of various clones was assessed using microtiter plates containing RNA samples from a variety of normal and pathology-derived cells, cell lines and tissues using real time quantitative PCR (RTQ PCR). RTQ PCR was performed on a Perkin-Elmer Biosystems ABI PRISM® 7700 Sequence Detection System. Various collections of samples are assembled on the plates, and referred to as Panel 1 (containing cells and cell lines from normal and cancer sources), Panel 2 (containing samples derived from tissues, in particular from surgical samples, from normal and cancer sources), Panel 3 (containing samples derived from a wide variety of cancer sources), Panel 4 (containing cells and cell lines from normal cells and cells related to inflammatory conditions) and Panel CNSD.01 (containing samples from normal and diseased brains).

First, the RNA samples were normalized to reference nucleic acids such as constitutively expressed genes (for example, β-actin and GAPDH). Normalized RNA (5 ul) was converted to cDNA and analyzed by RTQ-PCR using One Step RT-PCR Master Mix Reagents (PE Biosystems; Catalog No. 4309169) and gene-specific primers according to the manufacturer's instructions. Probes and primers were designed for each assay according to Perkin Elmer Biosystem's *Primer Express* Software package (version 1 for Apple Computer's Macintosh Power PC) or a similar algorithm using the target sequence as input. Default settings were used for reaction conditions and the following parameters were set before selecting primers: primer concentration = 250 nM, primer melting temperature (T_m) range = 58°-60° C, primer optimal Tm = 59° C, maximum primer difference = 2° C, probe does not have 5' G, probe T_m must be 10° C greater than primer T_m, amplicon size 75 bp to 100 bp. The probes and primers selected (see below) were synthesized by Synthegen (Houston, TX, USA). Probes were double purified by HPLC to remove uncoupled dye and evaluated by mass spectroscopy to verify coupling of reporter and quencher dyes to the 5' and 3' ends of the probe, respectively. Their final concentrations were: forward and reverse primers, 900 nM each, and probe, 200nM.

PCR conditions: Normalized RNA from each tissue and each cell line was spotted in each well of a 96 well PCR plate (Perkin Elmer Biosystems). PCR cocktails including two

probes (a probe specific for the target clone and another gene-specific probe multiplexed with the target probe) were set up using 1X TaqMan™ PCR Master Mix for the PE Biosystems 7700, with 5 mM MgCl2, dNTPs (dA, G, C, U at 1:1:1:2 ratios), 0.25 U/ml AmpliTaq Gold™ (PE Biosystems), and 0.4 U/μl RNase inhibitor, and 0.25 U/μl reverse transcriptase. Reverse transcription was performed at 48° C for 30 minutes followed by amplification/PCR cycles as follows: 95° C 10 min, then 40 cycles of 95° C for 15 seconds, 60° C for 1 minute. Results were recorded as CT values (cycle at which a given sample crosses a threshold level of fluorescence) using a log scale, with the difference in RNA concentration between a given sample and the sample with the lowest CT value being represented as 2 to the power of delta CT. The percent relative expression is then obtained by taking the reciprocal of this RNA difference and multiplying by 100.

In the results for Panel 1, the following abbreviations are used:
ca. = carcinoma,
* = established from metastasis,
met = metastasis,
s cell var = small cell variant,
non-s = non-sm = non-small,
squam = squamous,
pl. eff = pl effusion = pleural effusion,
glio = glioma,
astro = astrocytoma, and
neuro = neuroblastoma.

Panel 2

The plates for Panel 2 generally include 2 control wells and 94 test samples composed of RNA or cDNA isolated from human tissue procured by surgeons working in close cooperation with the National Cancer Institute's Cooperative Human Tissue Network (CHTN) or the National Disease Research Initiative (NDRI). The tissues are derived from human malignancies and in cases where indicated many malignant tissues have "matched margins" obtained from noncancerous tissue just adjacent to the tumor. These are termed normal adjacent tissues and are denoted "NAT" in the results below. The tumor tissue and the "matched margins" are evaluated by two independent pathologists (the surgical pathologists and again by a pathologists at NDRI or CHTN). This analysis provides a gross histopathological assessment of tumor differentiation grade. Moreover, most samples include the original surgical pathology report that provides information regarding the clinical stage of the patient. These matched margins are taken from the tissue surrounding (*i.e.* immediately proximal) to the zone of surgery (designated "NAT", for normal adjacent tissue, in Table RR). In addition, RNA and cDNA samples were obtained from various human tissues derived from autopsies performed on elderly people or sudden death

victims (accidents, etc.). These tissues were ascertained to be free of disease and were purchased from various commercial sources such as Clontech (Palo Alto, CA), Research Genetics, and Invitrogen.

RNA integrity from all samples is controlled for quality by visual assessment of agarose gel electropherograms using 28S and 18S ribosomal RNA staining intensity ratio as a guide (2:1 to 2.5:1 28s:18s) and the absence of low molecular weight RNAs that would be indicative of degradation products. Samples are controlled against genomic DNA contamination by RTQ PCR reactions run in the absence of reverse transcriptase using probe and primer sets designed to amplify across the span of a single exon.

Panel 3D

The plates of Panel 3D are comprised of 94 cDNA samples and two control samples. Specifically, 92 of these samples are derived from cultured human cancer cell lines, 2 samples of human primary cerebellar tissue and 2 controls. The human cell lines are generally obtained from ATCC (American Type Culture Collection), NCI or the German tumor cell bank and fall into the following tissue groups: Squamous cell carcinoma of the tongue, breast cancer, prostate cancer, melanoma, epidermoid carcinoma, sarcomas, bladder carcinomas, pancreatic cancers, kidney cancers, leukemias/lymphomas, ovarian/uterine/cervical, gastric, colon, lung and CNS cancer cell lines. In addition, there are two independent samples of cerebellum. These cells are all cultured under standard recommended conditions and RNA extracted using the standard procedures. The cell lines in panel 3D and 1.3D are of the most common cell lines used in the scientific literature.

RNA integrity from all samples is controlled for quality by visual assessment of agarose gel electropherograms using 28S and 18S ribosomal RNA staining intensity ratio as a guide (2:1 to 2.5:1 28s:18s) and the absence of low molecular weight RNAs that would be indicative of degradation products. Samples are controlled against genomic DNA contamination by RTQ PCR reactions run in the absence of reverse transcriptase using probe and primer sets designed to amplify across the span of a single exon.

Panel 4

Panel 4 includes samples on a 96 well plate (2 control wells, 94 test samples) composed of RNA (Panel 4r) or cDNA (Panel 4d) isolated from various human cell lines or tissues related to inflammatory conditions. Total RNA from control normal tissues such as colon and lung (Stratagene ,La Jolla, CA) and thymus and kidney (Clontech) were employed. Total RNA from liver tissue from cirrhosis patients and kidney from lupus patients was obtained from BioChain

(Biochain Institute, Inc., Hayward, CA). Intestinal tissue for RNA preparation from patients diagnosed as having Crohn's disease and ulcerative colitis was obtained from the National Disease Research Interchange (NDRI) (Philadelphia, PA).

Astrocytes, lung fibroblasts, dermal fibroblasts, coronary artery smooth muscle cells, small airway epithelium, bronchial epithelium, microvascular dermal endothelial cells, microvascular lung endothelial cells, human pulmonary aortic endothelial cells, human umbilical vein endothelial cells were all purchased from Clonetics (Walkersville, MD) and grown in the media supplied for these cell types by Clonetics. These primary cell types were activated with various cytokines or combinations of cytokines for 6 and/or 12-14 hours, as indicated. The following cytokines were used; IL-1 beta at approximately 1-5 ng/ml, TNF alpha at approximately 5-10 ng/ml, IFN gamma at approximately 20-50 ng/ml, IL-4 at approximately 5-10 ng/ml, IL-9 at approximately 5-10 ng/ml, IL-13 at approximately 5-10 ng/ml. Endothelial cells were sometimes starved for various times by culture in the basal media from Clonetics with 0.1% serum.

Mononuclear cells were prepared from blood of employees at CuraGen Corporation, using Ficoll. LAK cells were prepared from these cells by culture in DMEM 5% FCS (Hyclone), 100 μM non essential amino acids (Gibco/Life Technologies, Rockville, MD), 1 mM sodium pyruvate (Gibco), mercaptoethanol 5.5×10^{-5} M (Gibco), and 10 mM Hepes (Gibco) and Interleukin 2 for 4-6 days. Cells were then either activated with 10-20 ng/ml PMA and 1-2 μg/ml ionomycin, IL-12 at 5-10 ng/ml, IFN gamma at 20-50 ng/ml and IL-18 at 5-10 ng/ml for 6 hours. In some cases, mononuclear cells were cultured for 4-5 days in DMEM 5% FCS (Hyclone), 100 μM non essential amino acids (Gibco), 1 mM sodium pyruvate (Gibco), mercaptoethanol 5.5×10^{-5} M (Gibco), and 10 mM Hepes (Gibco) with PHA (phytohemagglutinin) or PWM (pokeweed mitogen) at approximately 5 μg/ml. Samples were taken at 24, 48 and 72 hours for RNA preparation. MLR (mixed lymphocyte reaction) samples were obtained by taking blood from two donors, isolating the mononuclear cells using Ficoll and mixing the isolated mononuclear cells 1:1 at a final concentration of approximately 2×10^6 cells/ml in DMEM 5% FCS (Hyclone), 100 μM non essential amino acids (Gibco), 1 mM sodium pyruvate (Gibco), mercaptoethanol (5.5×10^{-5} M) (Gibco), and 10 mM Hepes (Gibco). The MLR was cultured and samples taken at various time points ranging from 1- 7 days for RNA preparation.

Monocytes were isolated from mononuclear cells using CD14 Miltenyi Beads, **+ve** VS selection columns and a Vario Magnet according to the manufacturer's instructions. Monocytes were differentiated into dendritic cells by culture in DMEM 5% fetal calf serum (FCS) (Hyclone, Logan, UT), 100 μM non essential amino acids (Gibco), 1 mM sodium pyruvate

(Gibco), mercaptoethanol 5.5 x 10^{-5} M (Gibco), and 10 mM Hepes (Gibco), 50 ng/ml GMCSF and 5 ng/ml IL-4 for 5-7 days. Macrophages were prepared by culture of monocytes for 5-7 days in DMEM 5% FCS (Hyclone), 100 μM non essential amino acids (Gibco), 1 mM sodium pyruvate (Gibco), mercaptoethanol 5.5 x 10^{-5} M (Gibco), 10 mM Hepes (Gibco) and 10% AB Human Serum or MCSF at approximately 50 ng/ml. Monocytes, macrophages and dendritic cells were stimulated for 6 and 12-14 hours with lipopolysaccharide (LPS) at 100 ng/ml. Dendritic cells were also stimulated with anti-CD40 monoclonal antibody (Pharmingen) at 10 μg/ml for 6 and 12-14 hours.

CD4 lymphocytes, CD8 lymphocytes and NK cells were also isolated from mononuclear cells using CD4, CD8 and CD56 Miltenyi beads. positive VS selection columns and a Vario Magnet according to the manufacturer's instructions. CD45RA and CD45RO CD4 lymphocytes were isolated by depleting mononuclear cells of CD8, CD56, CD14 and CD19 cells using CD8, CD56, CD14 and CD19 Miltenyi beads and positive selection. Then CD45RO beads were used to isolate the CD45RO CD4 lymphocytes with the remaining cells being CD45RA CD4 lymphocytes. CD45RA CD4, CD45RO CD4 and CD8 lymphocytes were placed in DMEM 5% FCS (Hyclone), 100 μM non essential amino acids (Gibco), 1 mM sodium pyruvate (Gibco), mercaptoethanol 5.5 x 10^{-5} M (Gibco). and 10 mM Hepes (Gibco) and plated at 10^6 cells/ml onto Falcon 6 well tissue culture plates that had been coated overnight with 0.5 μg/ml anti-CD28 (Pharmingen) and 3 ug/ml anti-CD3 (OKT3, ATCC) in PBS. After 6 and 24 hours, the cells were harvested for RNA preparation. To prepare chronically activated CD8 lymphocytes, we activated the isolated CD8 lymphocytes for 4 days on anti-CD28 and anti-CD3 coated plates and then harvested the cells and expanded them in DMEM 5% FCS (Hyclone), 100 μM non essential amino acids (Gibco), 1 mM sodium pyruvate (Gibco), mercaptoethanol 5.5 x 10^{-5} M (Gibco), and 10 mM Hepes (Gibco) and IL-2. The expanded CD8 cells were then activated again with plate bound anti-CD3 and anti-CD28 for 4 days and expanded as before. RNA was isolated 6 and 24 hours after the second activation and after 4 days of the second expansion culture. The isolated NK cells were cultured in DMEM 5% FCS (Hyclone), 100 μM non essential amino acids (Gibco), 1 mM sodium pyruvate (Gibco), mercaptoethanol 5.5 x 10^{-5} M (Gibco), and 10 mM Hepes (Gibco) and IL-2 for 4-6 days before RNA was prepared.

To obtain B cells, tonsils were procured from NDRI. The tonsil was cut up with sterile dissecting scissors and then passed through a sieve. Tonsil cells were then spun down and resupended at 10^6 cells/ml in DMEM 5% FCS (Hyclone), 100 μM non essential amino acids (Gibco), 1 mM sodium pyruvate (Gibco), mercaptoethanol 5.5 x 10^{-5} M (Gibco), and 10 mM Hepes (Gibco). To activate the cells, we used PWM at 5 μg/ml or anti-CD40 (Pharmingen) at

approximately 10 μg/ml and IL-4 at 5-10 ng/ml. Cells were harvested for RNA preparation at 24,48 and 72 hours.

To prepare the primary and secondary Th1/Th2 and Tr1 cells, six-well Falcon plates were coated overnight with 10 μg/ml anti-CD28 (Pharmingen) and 2 μg/ml OKT3 (ATCC), and then washed twice with PBS. Umbilical cord blood CD4 lymphocytes (Poietic Systems, German Town, MD) were cultured at 10^5-10^6 cells/ml in DMEM 5% FCS (Hyclone), 100 μM non essential amino acids (Gibco), 1 mM sodium pyruvate (Gibco), mercaptoethanol 5.5 x 10^{-5} M (Gibco), 10 mM Hepes (Gibco) and IL-2 (4 ng/ml). IL-12 (5 ng/ml) and anti-IL4 (1 μg/ml) were used to direct to Th1, while IL-4 (5 ng/ml) and anti-IFN gamma (1 μg/ml) were used to direct to Th2 and IL-10 at 5 ng/ml was used to direct to Tr1. After 4-5 days, the activated Th1, Th2 and Tr1 lymphocytes were washed once in DMEM and expanded for 4-7 days in DMEM 5% FCS (Hyclone), 100 μM non essential amino acids (Gibco), 1 mM sodium pyruvate (Gibco), mercaptoethanol 5.5 x 10^{-5} M (Gibco), 10 mM Hepes (Gibco) and IL-2 (1 ng/ml). Following this, the activated Th1, Th2 and Tr1 lymphocytes were re-stimulated for 5 days with anti-CD28/OKT3 and cytokines as described above, but with the addition of anti-CD95L (1 μg/ml) to prevent apoptosis. After 4-5 days, the Th1, Th2 and Tr1 lymphocytes were washed and then expanded again with IL-2 for 4-7 days. Activated Th1 and Th2 lymphocytes were maintained in this way for a maximum of three cycles. RNA was prepared from primary and secondary Th1, Th2 and Tr1 after 6 and 24 hours following the second and third activations with plate bound anti-CD3 and anti-CD28 mAbs and 4 days into the second and third expansion cultures in Interleukin 2.

The following leukocyte cells lines were obtained from the ATCC: Ramos, EOL-1, KU-812. EOL cells were further differentiated by culture in 0.1 mM dbcAMP at 5 x10^5 cells/ml for 8 days, changing the media every 3 days and adjusting the cell concentration to 5 x10^5 cells/ml. For the culture of these cells, we used DMEM or RPMI (as recommended by the ATCC), with the addition of 5% FCS (Hyclone), 100 μM non essential amino acids (Gibco), 1 mM sodium pyruvate (Gibco), mercaptoethanol 5.5 x 10^{-5} M (Gibco), 10 mM Hepes (Gibco). RNA was either prepared from resting cells or cells activated with PMA at 10 ng/ml and ionomycin at 1 μg/ml for 6 and 14 hours. Keratinocyte line CCD106 and an airway epithelial tumor line NCI-H292 were also obtained from the ATCC. Both were cultured in DMEM 5% FCS (Hyclone), 100 μM non essential amino acids (Gibco), 1 mM sodium pyruvate (Gibco), mercaptoethanol 5.5 x 10^{-5} M (Gibco), and 10 mM Hepes (Gibco). CCD1106 cells were activated for 6 and 14 hours with approximately 5 ng/ml TNF alpha and 1 ng/ml IL-1 beta, while NCI-H292 cells were

activated for 6 and 14 hours with the following cytokines: 5 ng/ml IL-4, 5 ng/ml IL-9, 5 ng/ml IL-13 and 25 ng/ml IFN gamma.

For these cell lines and blood cells, RNA was prepared by lysing approximately 10^7 cells/ml using Trizol (Gibco BRL). Briefly, 1/10 volume of bromochloropropane (Molecular Research Corporation) was added to the RNA sample, vortexed and after 10 minutes at room temperature, the tubes were spun at 14,000 rpm in a Sorvall SS34 rotor. The aqueous phase was removed and placed in a 15 ml Falcon Tube. An equal volume of isopropanol was added and left at –20 degrees C overnight. The precipitated RNA was spun down at 9,000 rpm for 15 min in a Sorvall SS34 rotor and washed in 70% ethanol. The pellet was redissolved in 300 µl of RNAse-free water and 35 µl buffer (Promega) 5 µl DTT, 7 µl RNAsin and 8 µl DNAse were added. The tube was incubated at 37 degrees C for 30 minutes to remove contaminating genomic DNA, extracted once with phenol chloroform and re-precipitated with 1/10 volume of 3 M sodium acetate and 2 volumes of 100% ethanol. The RNA was spun down and placed in RNAse free water. RNA was stored at –80 degrees C.

Panel CNSD.01

The plates for Panel CNSD.01 include two control wells and 94 test samples comprised of cDNA isolated from postmortem human brain tissue obtained from the Harvard Brain Tissue Resource Center. Brains are removed from calvaria of donors between 4 and 24 hours after death, sectioned by neuroanatomists, and frozen at -80°C in liquid nitrogen vapor. All brains are sectioned and examined by neuropathologists to confirm diagnoses with clear associated neuropathology.

Disease diagnoses are taken from patient records. The panel contains two brains from each of the following diagnoses: Alzheimer's disease, Parkinson's disease, Huntington's disease, Progressive Supernuclear Palsy, Depression, and "Normal controls". Within each of these brains, the following regions are represented: cingulate gyrus, temporal pole, globus palladus, substantia nigra, Brodman Area 4 (primary motor strip), Brodman Area 7 (parietal cortex), Brodman Area 9 (prefrontal cortex), and Brodman area 17 (occipital cortex). Not all brain regions are represented in all cases; *e.g.*, Huntington's disease is characterized in part by neurodegeneration in the globus palladus, thus this region is impossible to obtain from confirmed Huntington's cases. Likewise Parkinson's disease is characterized by degeneration of the substantia nigra making this region more difficult to obtain. Normal control brains were examined for neuropathology and found to be free of any pathology consistent with neurodegeneration.

RNA integrity from all samples is controlled for quality by visual assessment of agarose gel electropherograms using 28S and 18S ribosomal RNA staining intensity ratio as a guide (2:1 to 2.5:1 28s:18s) and the absence of low molecular weight RNAs that would be indicative of degradation products. Samples are controlled against genomic DNA contamination by RTQ PCR reactions run in the absence of reverse transcriptase using probe and primer sets designed to amplify across the span of a single exon.

In the labels employed to identify tissues in the CNS panel, the following abbreviations are used:

PSP = Progressive supranuclear palsy

Sub Nigra = Substantia nigra

Glob Palladus= Globus palladus

Temp Pole = Temporal pole

Cing Gyr = Cingulate gyrus

BA 4 = Brodman Area 4

The AC068339_A gene encodes a G protein-coupled receptor (GPCR), a type of cell surface receptor involved in signal transduction. The AC068339_A gene product is most similar to members of the odorant receptor subfamily of GPCRs. Based on analogy to other odorant receptor genes, we predict that expression of the AC068339_A gene may be highest in nasal epithelium, a sample not represented on these panels.

NOV1 - 24CS059

Expression of the NOV1 gene, referred to as 24CS059, was assessed using the primer-probe set Ag3975, described in Table 18A. Results from RTQ-PCR runs are shown in Tables 18B and 18C.

Table 18A. Probe Name Ag3975

Primers	Sequences	TM	Length	Start Position	SEQ ID NO
Forward	5'-CTGAACTCAGTTGGCAAAGG-3'	58.5	20	5598	124
Probe	FAM-5'-TCTGTGGGTAAATCCTCTTTCACATG-3'-TAMRA	64.5	26	5622	125
Reverse	5'-AGGGCCACATCATGTATGTTAG-3'	58.9	22	5672	126

Table 18B. Panel 2.1

Tissue Name	Relative Expression(%) 2.1x4tm6080f_ag3975_a1	Tissue Name	Relative Expression(%) 2.1x4tm6080f_ag3975_a1
Normal Colon GENPAK 061003	41.7	Kidney Cancer Clontech 9010320	0.0

97759 Colon cancer (OD06064)	7.2	Kidney NAT Clontech 9010321	52.9
97760 Colon cancer NAT (OD06064)	0.0	Kidney Cancer Clontech 8120607	3.3
97778 Colon cancer (OD06159)	0.0	Kidney NAT Clontech 8120608	0.0
97779 Colon cancer NAT (OD06159)	10.2	Normal Uterus GENPAK 061018	33.6
98859 Colon cancer (OD06298-08)	5.3	Uterus Cancer GENPAK 064011	10.8
98860 Colon cancer NAT (OD06298-018)	0.0	Normal Thyroid Clontech A+ 6570-1 (7080817)	8.3
83237 CC Gr.2 ascend colon (OD03921)	41.1	Thyroid Cancer GENPAK 064010	22.6
83238 CC NAT (OD03921)	30.9	Thyroid Cancer INVITROGEN A302152	44.3
97766 Colon cancer metastasis (OD06104)	17.0	Thyroid NAT INVITROGEN A302153	82.8
97767 Lung NAT (OD06104)	0.0	Normal Breast GENPAK 061019	88.8
87472 Colon mets to lung (OD04451-01)	23.5	84877 Breast Cancer (OD04566)	40.5
87473 Lung NAT (OD04451-02)	12.0	Breast Cancer Res. Gen. 1024	18.7
Normal Prostate Clontech A+ 6546-1 (8090438)	2.5	85975 Breast Cancer (OD04590-01)	0.0
84140 Prostate Cancer (OD04410)	4.5	85976 Breast Cancer Mets (OD04590-03)	17.9
84141 Prostate NAT (OD04410)	8.1	87070 Breast Cancer Metastasis (OD04655-05)	10.8
Normal Lung GENPAK 061010	28.0	GENPAK Breast Cancer 064006	10.2
92337 Invasive poor diff. lung adeno (OD04945-01	26.1	Breast Cancer Clontech 9100266	13.6
92338 Lung NAT (OD04945-03)	99.1	Breast NAT Clontech 9100265	34.7
84136 Lung Malignant Cancer (OD03126)	4.3	Breast Cancer INVITROGEN A209073	0.0
84137 Lung NAT (OD03126)	30.4	Breast NAT INVITROGEN A2090734	79.2
90372 Lung Cancer (OD05014A)	19.9	Normal Liver GENPAK 061009	44.5
90373 Lung NAT (OD05014B)	30.8	Liver Cancer Research Genetics RNA 1026	0.0
85950 Lung Cancer (OD04237-01)	18.1	Liver Cancer Research Genetics RNA 1025	8.3
85970 Lung NAT (OD04237-02)	42.7	Paired Liver Cancer Tissue Research Genetics RNA 6004-T	4.2
83255 Ocular Mel Met to Liver (OD04310)	18.6	Paired Liver Tissue Research Genetics RNA 6004-N	0.0
83256 Liver NAT (OD04310)	11.2	Paired Liver Cancer Tissue Research Genetics RNA 6005-T	3.7
84139 Melanoma Mets to Lung (OD04321)	32.4	Paired Liver Tissue Research Genetics RNA 6005-N	0.0
84138 Lung NAT (OD04321)	13.8	Liver Cancer GENPAK 064003	0.0
Normal Kidney GENPAK 061008	44.0	Normal Bladder GENPAK 061001	35.0
83786 Kidney Ca, Nuclear grade 2 (OD04338)	41.7	Bladder Cancer Research Genetics RNA 1023	0.0
83787 Kidney NAT (OD04338)	51.4	Bladder Cancer INVITROGEN A302173	36.6
83788 Kidney Ca Nuclear grade 1/2 (OD04339)	22.1	Normal Ovary Res. Gen.	0.0

83789 Kidney NAT (OD04339)	10.5	Ovarian Cancer GENPAK 064008	2.9
83790 Kidney Ca, Clear cell type (OD04340)	20.1	97773 Ovarian cancer (OD06145)	0.0
83791 Kidney NAT (OD04340)	34.2	97775 Ovarian cancer NAT (OD06145)	9.9
83792 Kidney Ca, Nuclear grade 3 (OD04348)	4.6	Normal Stomach GENPAK 061017	14.6
83793 Kidney NAT (OD04348)	17.4	Gastric Cancer Clontech 9060397	0.0
85973 Kidney Cancer (OD04450-01)	100.0	NAT Stomach Clontech 9060396	6.3
85974 Kidney NAT (OD04450-03)	55.5	Gastric Cancer Clontech 9060395	39.4
Kidney Cancer Clontech 8120613	0.0	NAT Stomach Clontech 9060394	16.6
Kidney NAT Clontech 8120614	0.0	Gastric Cancer GENPAK 064005	55.3

Table 18C. Panel 4.1D

Tissue Name	Relative Expression(%) 4.1dx4tm6081f_ag3975_a1	Tissue Name	Relative Expression(%) 4.1dx4tm6081f_ag3975_a1
93768_Secondary Th1_anti-CD28/anti-CD3	3.2	93100_HUVEC (Endothelial)_IL-1b	0.6
93769_Secondary Th2_anti-CD28/anti-CD3	4.3	93779_HUVEC (Endothelial)_IFN gamma	0.3
93770_Secondary Tr1_anti-CD28/anti-CD3	2.0	93102_HUVEC (Endothelial)_TNF alpha + IFN gamma	1.7
93573_Secondary Th1_resting day 4-6 in IL-2	0.0	93101_HUVEC (Endothelial)_TNF alpha + IL4	1.3
93572_Secondary Th2_resting day 4-6 in IL-2	0.8	93781_HUVEC (Endothelial)_IL-11	0.5
93571_Secondary Tr1_resting day 4-6 in IL-2	0.0	93583_Lung Microvascular Endothelial Cells_none	3.2
93568_primary Th1_anti-CD28/anti-CD3	4.3	93584_Lung Microvascular Endothelial Cells_TNFa (4 ng/ml) and IL1b (1 ng/ml)	4.2
93569_primary Th2_anti-CD28/anti-CD3	6.0	92662_Microvascular Dermal endothelium_none	1.1
93570_primary Tr1_anti-CD28/anti-CD3	2.8	92663_Microsvasular Dermal endothelium_TNFa (4 ng/ml) and IL1b (1 ng/ml)	1.4
93565_primary Th1_resting dy 4-6 in IL-2	0.0	93773_Bronchial epithelium_TNFa (4 ng/ml) and IL1b (1 ng/ml) **	5.8
93566_primary Th2_resting dy 4-6 in IL-2	0.4	93347_Small Airway Epithelium_none	0.0
93567_primary Tr1_resting dy 4-6 in IL-2	0.3	93348_Small Airway Epithelium_TNFa (4 ng/ml) and IL1b (1 ng/ml)	6.3
93351_CD45RA CD4 lymphocyte_anti-CD28/anti-CD3	3.0	92668_Coronery Artery SMC_resting	1.4
93352_CD45RO CD4 lymphocyte_anti-CD28/anti-CD3	3.2	92669_Coronery Artery SMC_TNFa (4 ng/ml) and IL1b (1 ng/ml)	0.2
93251_CD8 Lymphocytes_anti-CD28/anti-CD3	0.9	93107_astrocytes_resting	1.0

93353_chronic CD8 Lymphocytes 2ry_resting dy 4-6 in IL-2	4.9	93108_astrocytes_TNFa (4 ng/ml) and ILlb (1 ng/ml)	1.8
93574_chronic CD8 Lymphocytes 2ry_activated CD3/CD28	0.6	92666_KU-812 (Basophil)_resting	3.3
93354_CD4_none	0.4	92667_KU-812 (Basophil)_PMA/ionoycin	3.9
93252_Secondary Th1/Th2/Tr1_anti-CD95 CH11	0.8	93579_CCD1106 (Keratinocytes)_none	6.1
93103_LAK cells_resting	1.2	93580_CCD1106 (Keratinocytes)_TNFa and IFNg **	10.2
93788_LAK cells_IL-2	1.2	93791_Liver Cirrhosis	4.2
93787_LAK cells_IL-2+IL-12	100.0	93577_NCI-H292	17.7
93789_LAK cells_IL-2+IFN gamma	1.8	93358_NCI-H292_IL-4	18.5
93790_LAK cells_IL-2+ IL-18	3.8	93360_NCI-H292_IL-9	19.4
93104_LAK cells_PMA/ionomycin and IL-18	3.1	93359_NCI-H292_IL-13	13.3
93578_NK Cells IL-2_resting	1.1	93357_NCI-H292_IFN gamma	14.2
93109_Mixed Lymphocyte Reaction_Two Way MLR	3.9	93777_HPAEC_-	0.0
93110_Mixed Lymphocyte Reaction_Two Way MLR	2.2	93778_HPAEC_IL-1 beta/TNA alpha	2.9
93111_Mixed Lymphocyte Reaction_Two Way MLR	0.7	93254_Normal Human Lung Fibroblast_none	1.0
93112_Mononuclear Cells (PBMCs)_resting	0.3	93253_Normal Human Lung Fibroblast_TNFa (4 ng/ml) and IL-1b (1 ng/ml)	0.5
93113_Mononuclear Cells (PBMCs)_PWM	6.8	93257_Normal Human Lung Fibroblast_IL-4	0.4
93114_Mononuclear Cells (PBMCs)_PHA-L	2.0	93256_Normal Human Lung Fibroblast_IL-9	1.2
93249_Ramos (B cell)_none	31.1	93255_Normal Human Lung Fibroblast_IL-13	0.7
93250_Ramos (B cell)_ionomycin	34.8	93258_Normal Human Lung Fibroblast_IFN gamma	1.4
93349_B lymphocytes_PWM	5.9	93106_Dermal Fibroblasts CCD1070_resting	0.8
93350_B lymphoytes_CD40L and IL-4	11.2	93361_Dermal Fibroblasts CCD1070_TNF alpha 4 ng/ml	1.9
92665_EOL-1 (Eosinophil)_dbcAMP differentiated	13.0	93105_Dermal Fibroblasts CCD1070_IL-1 beta 1 ng/ml	0.0
93248_EOL-1 (Eosinophil)_dbcAMP/PMAionomycin	3.6	93772_dermal fibroblast_IFN gamma	0.3
93356_Dendritic Cells_none	4.0	93771_dermal fibroblast_IL-4	1.0
93355_Dendritic Cells_LPS 100 ng/ml	2.2	93892_Dermal fibroblasts_none	0.7
93775_Dendritic Cells_anti-CD40	4.1	99202_Neutrophils_TNFa+LPS	7.3
93774_Monocytes_resting	7.4	99203_Neutrophils_none	8.3
93776_Monocytes_LPS 50 ng/ml	11.4	735010_Colon_normal	3.0
93581_Macrophages_resting	0.8	735019_Lung_none	4.8
93582_Macrophages_LPS 100 ng/ml	0.0	64028-1_Thymus_none	19.9
93098_HUVEC (Endothelial)_none	0.9	64030-1_Kidney_none	50.8

93099_HUVEC (Endothelial) starved	0.4		

Panel 2.1 Summary: Ag3975 The level of expression of the NOV1 - 24CS059 gene is low in the samples used for Panel 2.1, with highest expression in a kidney cancer sample (CT = 33.9). However, expression of this gene shows a moderate association with samples derived from gastric cancer when compared to their associated normal adjacent tissue as well as with a single sample of renal cancer compared with normal adjacent tissue. Thus, based upon its profile, the expression of the 24CS059 gene could be of use as a marker for gastric cancer. In addition, therapeutic inhibition of the activity of this gene product, through the use of antibodies or small molecule drugs, may be useful in the therapy of gastric cancer.

Panel 4.1D Summary: Ag3975 The NOV1 - 24CS059 gene is most highly expressed in LAK cells activated by treatment with IL-2 and IL-12 (CT = 29.5). This expression appears to be induced by IL-12 treatment since LAK treated with only IL-2 shows is expressed at much lower levels (CT = 35.9). IL-12 has been shown to synergize with IL-2 to augment NK- and induce LAK-mediated cytotoxicity; this synergistic increase is associated with enhanced transcription of perforin and granzyme genes (ref. 1). Activated LAK cells are able to lyse a wide range of targets including fresh tumor cells and virally infected cells. Therefore, the NOV1 protein encoded by the 24CS059 gene could be used as a protein therapeutic in the treatment of many cancerous tumors and also in infectious disease, (viral disease in particular). Additional low but significant expression of the 24CS059 gene is seen in activated B cells, in a mucoepidermoid carcinoma cell line and in monocytes but not on macrophages, suggesting that this protein is down regulated during macrophage differentiation.

References:

(1). DeBlaker-Hohe D.F., Yamauchi A., Yu C.R., Horvath-Arcidiacono J.A., Bloom E.T. (1995) IL-12 synergizes with IL-2 to induce lymphokine-activated cytotoxicity and perforin and granzyme gene expression in fresh human NK cells. Cell. Immunol. 165: 33-43.

NK-mediated cytotoxicity is regulated by a variety of cytokines and is thought to involve perforin and granzymes. The effects of IL-2 and IL-12 on the expression and activation of cytolysis were examined in freshly isolated human NK cells. A dose-dependent increase in cytolysis of the NK-sensitive target cell, K562, and the NK-insensitive but lymphokine-activated killer (LAK) cell-sensitive target, UCLA-SO-M14, was observed after short term culture of purified human NK cells in either IL-2 or IL-12. Moreover, the two cytokines often synergized to produce augmented lytic activity. A suboptimal dose of IL-2 (60 IU/ml) combined with IL-12 (2 U/ml) could induce lytic activity equal to twice the additive effect of each cytokine alone.

Northern analyses revealed time-dependent increases in mRNAs encoding for perforin and granzymes A and B following treatment with IL-2 alone or IL-2 plus IL-12. IL-2 and IL-12 also synergized for the induction of granzyme mRNAs, in that treatment with both cytokines increased mRNA levels approximately 50% above the sum of each cytokine alone, as quantitated by phosphorimage analysis, and normalized to GAPDH gene expression. However, the synergy between IL-2 and IL-12 for the induction of mRNA was less dramatic than for lytic activity. Results of experiments in which cytokine-treated cells were pulsed with actinomycin D indicated that the increased granzyme and perforin gene mRNA levels in response to IL-2, IL-12, or the combination were not due to increased transcript stability. The data suggest that low doses of IL-2 and IL-12 synergize to augment NK- and induce LAK-mediated cytotoxicity and that this increase is associated with enhanced transcription of perforin and granzyme genes in a synergistic fashion. PMID: 7671323

NOV3 - 24SC113

Expression of gene 24SC113 was assessed using the primer-probe set Ag1460, described in Table 19A. Results from RTQ-PCR runs are shown in Tables 19B and 19C.

Table 19A. Probe Name Ag1460

Primers	Sequences	TM	Length	Start Position	SEQ ID NO
Forward	5'-CCCTGAAATACACAGAGGACAT-3'	58.1	22	860	125
Probe	FAM-5'-ATGGAATCCCTGGCCCTGTCTAATG-3'-TAMRA	68.9	25	913	126
Reverse	5'-GGTGAACAGAACCTACCTGTTG-3'	58.6	22	938	127

Table 19B. Panel 2.1

Tissue Name	Relative Expression(%) 2.1tm6078f_ag1460	Tissue Name	Relative Expression(%) 2.1tm6078f_ag1460
Normal Colon GENPAK 061003	8.0	Kidney Cancer Clontech 9010320	3.2
97759 Colon cancer (OD06064)	8.7	Kidney NAT Clontech 9010321	55.5
97760 Colon cancer NAT (OD06064)	0.0	Kidney Cancer Clontech 8120607	2.3
97778 Colon cancer (OD06159)	0.0	Kidney NAT Clontech 8120608	7.3
97779 Colon cancer NAT (OD06159)	6.7	Normal Uterus GENPAK 061018	100.0
98859 Colon cancer (OD06298-08)	6.9	Uterus Cancer GENPAK 064011	23.0
98860 Colon cancer NAT (OD06298-018)	3.6	Normal Thyroid Clontech A+ 6570-1 (7080817)	3.7
83237 CC Gr.2 ascend colon (OD03921)	2.8	Thyroid Cancer GENPAK 064010	0.0
83238 CC NAT (OD03921)	12.0	Thyroid Cancer INVITROGEN	6.7

		A302152	
97766 Colon cancer metastasis (OD06104)	4.1	Thyroid NAT INVITROGEN A302153	29.1
97767 Lung NAT (OD06104)	3.6	Normal Breast GENPAK 061019	20.0
84872 Colon mets to lung (OD04451-01)	0.0	84877 Breast Cancer (OD04566)	12.2
84873 Lung NAT (OD04451-02)	28.3	Breast Cancer Res. Gen. 1024	30.8
Normal Prostate Clontech A+ 6546-1 (8090438)	0.0	85975 Breast Cancer (OD04590-01)	4 1
84140 Prostate Cancer (OD04410)	0.0	85976 Breast Cancer Mets (OD04590-03)	16.3
84141 Prostate NAT (OD04410)	0.0	87070 Breast Cancer Metastasis (OD04655-05)	26.8
Normal Lung GENPAK 061010	53.6	GENPAK Breast Cancer 064006	3 2
92337 Invasive poor diff. lung adeno (OD04945-01	4.5	Breast Cancer Clontech 9100266	2.0
92338 Lung NAT (OD04945-03)	71.2	Breast NAT Clontech 9100265	8.7
84136 Lung Malignant Cancer (OD03126)	5.6	Breast Cancer INVITROGEN A209073	8 8
84137 Lung NAT (OD03126)	10.4	Breast NAT INVITROGEN A2090734	32.3
90372 Lung Cancer (OD05014A)	7.3	Normal Liver GENPAK 061009	5.6
90373 Lung NAT (OD05014B)	19.5	Liver Cancer Research Genetics RNA 1026	0 0
85950 Lung Cancer (OD04237-01)	11.0	Liver Cancer Research Genetics RNA 1025	5 0
85970 Lung NAT (OD04237-02)	19.6	Paired Liver Cancer Tissue Research Genetics RNA 6004-T	18.2
83255 Ocular Mel Met to Liver (OD04310)	0.0	Paired Liver Tissue Research Genetics RNA 6004-N	0 0
83256 Liver NAT (OD04310)	10.2	Paired Liver Cancer Tissue Research Genetics RNA 6005-T	3.7
84139 Melanoma Mets to Lung (OD04321)	7.9	Paired Liver Tissue Research Genetics RNA 6005-N	7.5
84138 Lung NAT (OD04321)	11.6	Liver Cancer GENPAK 064003	0.0
Normal Kidney GENPAK 061008	15.0	Normal Bladder GENPAK 061001	3.0
83786 Kidney Ca, Nuclear grade 2 (OD04338)	15.4	Bladder Cancer Research Genetics RNA 1023	2.7
83787 Kidney NAT (OD04338)	12.9	Bladder Cancer INVITROGEN A302173	4.3
83788 Kidney Ca Nuclear grade 1/2 (OD04339)	12.5	Normal Ovary Res. Gen.	2.9
83789 Kidney NAT (OD04339)	8.5	Ovarian Cancer GENPAK 064008	22.5
83790 Kidney Ca, Clear cell type (OD04340)	3.4	97773 Ovarian cancer (OD06145)	0.0
83791 Kidney NAT (OD04340)	9.2	97775 Ovarian cancer NAT (OD06145)	33.2
83792 Kidney Ca, Nuclear grade 3 (OD04348)	0.0	Normal Stomach GENPAK 061017	65.5
83793 Kidney NAT (OD04348)	10.5	Gastric Cancer Clontech 9060397	3.6
85973 Kidney Cancer (OD04450-01)	29.9	NAT Stomach Clontech 9060396	0.0
85974 Kidney NAT (OD04450-03)	11.6	Gastric Cancer Clontech 9060395	17.3

Kidney Cancer Clontech 8120613	0.0	NAT Stomach Clontech 9060394	7.4
Kidney NAT Clontech 8120614	2.8	Gastric Cancer GENPAK 064005	2.5

Table 19C. Panel 4.1D

Tissue Name	Relative Expression(%) 4.1dx4tm5965f_ag1460_a1	Tissue Name	Relative Expression(%) 4.1dx4tm5965f_ag1460_a1
93768_Secondary Th1_anti-CD28/anti-CD3	0.0	93100_HUVEC (Endothelial)_IL-1b	4.7
93769_Secondary Th2_anti-CD28/anti-CD3	0.0	93779_HUVEC (Endothelial)_IFN gamma	9.3
93770_Secondary Tr1_anti-CD28/anti-CD3	1.3	93102_HUVEC (Endothelial)_TNF alpha + IFN gamma	4.1
93573_Secondary Th1_resting day 4-6 in IL-2	0.0	93101_HUVEC (Endothelial)_TNF alpha + IL4	3.0
93572_Secondary Th2_resting day 4-6 in IL-2	0.0	93781_HUVEC (Endothelial)_IL-11	9.0
93571_Secondary Tr1_resting day 4-6 in IL-2	0.0	93583_Lung Microvascular Endothelial Cells_none	2.5
93568_primary Th1_anti-CD28/anti-CD3	0.0	93584_Lung Microvascular Endothelial Cells_TNFa (4 ng/ml) and IL1b (1 ng/ml)	4.9
93569_primary Th2_anti-CD28/anti-CD3	0.0	92662_Microvascular Dermal endothelium_none	3.4
93570_primary Tr1_anti-CD28/anti-CD3	0.0	92663_Microsvasular Dermal endothelium_TNFa (4 ng/ml) and IL1b (1 ng/ml)	1.3
93565_primary Th1_resting dy 4-6 in IL-2	0.0	93773_Bronchial epithelium_TNFa (4 ng/ml) and IL1b (1 ng/ml) **	10.7
93566_primary Th2_resting dy 4-6 in IL-2	0.0	93347_Small Airway Epithelium_none	5.5
93567_primary Tr1_resting dy 4-6 in IL-2	0.0	93348_Small Airway Epithelium_TNFa (4 ng/ml) and IL1b (1 ng/ml)	22.2
93351_CD45RA CD4 lymphocyte_anti-CD28/anti-CD3	5.8	92668_Coronery Artery SMC_resting	12.3
93352_CD45RO CD4 lymphocyte_anti-CD28/anti-CD3	0.0	92669_Coronery Artery SMC_TNFa (4 ng/ml) and IL1b (1 ng/ml)	12.4
93251_CD8 Lymphocytes_anti-CD28/anti-CD3	0.0	93107_astrocytes_resting	100.0
93353_chronic CD8 Lymphocytes 2ry_resting dy 4-6 in IL-2	0.0	93108_astrocytes_TNFa (4 ng/ml) and IL1b (1 ng/ml)	15.4
93574_chronic CD8 Lymphocytes 2ry_activated CD3/CD28	0.0	92666_KU-812 (Basophil)_resting	0.0
93354_CD4_none	0.0	92667_KU-812 (Basophil)_PMA/ionoycin	0.0
93252_Secondary Th1/Th2/Tr1_anti-CD95 CH11	0.0	93579_CCD1106 (Keratinocytes)_none	16.5
93103_LAK cells_resting	0.0	93580_CCD1106 (Keratinocytes)_TNFa and IFNg **	16.8
93788_LAK cells_IL-2	0.0	93791_Liver Cirrhosis	4.7

93787_LAK cells_IL-2+IL-12	0.0	93577_NCI-H292	3.8
93789_LAK cells_IL-2+IFN gamma	0.0	93358_NCI-H292_IL-4	1.1
93790_LAK cells_IL-2+ IL-18	0.0	93360_NCI-H292_IL-9	5.7
93104_LAK cells_PMA/ionomycin and IL-18	0.0	93359_NCI-H292_IL-13	3.5
93578_NK Cells IL-2_resting	1.1	93357_NCI-H292_IFN gamma	1.5
93109_Mixed Lymphocyte Reaction_Two Way MLR	0.0	93777_HPAEC_-	10.6
93110_Mixed Lymphocyte Reaction_Two Way MLR	0.0	93778_HPAEC_IL-1 beta/TNA alpha	16.8
93111_Mixed Lymphocyte Reaction_Two Way MLR	0.0	93254_Normal Human Lung Fibroblast_none	12.7
93112_Mononuclear Cells (PBMCs)_resting	0.0	93253_Normal Human Lung Fibroblast_TNFa (4 ng/ml) and IL-1b (1 ng/ml)	8.7
93113_Mononuclear Cells (PBMCs)_PWM	0.0	93257_Normal Human Lung Fibroblast_IL-4	11.2
93114_Mononuclear Cells (PBMCs)_PHA-L	0.0	93256_Normal Human Lung Fibroblast_IL-9	23.6
93249_Ramos (B cell)_none	0.0	93255_Normal Human Lung Fibroblast_IL-13	27.2
93250_Ramos (B cell)_ionomycin	0.0	93258_Normal Human Lung Fibroblast_IFN gamma	13.3
93349_B lymphocytes_PWM	0.0	93106_Dermal Fibroblasts CCD1070_resting	27.1
93350_B lymphoytes_CD40L and IL-4	1.4	93361_Dermal Fibroblasts CCD1070_TNF alpha 4 ng/ml	1.2
92665_EOL-1 (Eosinophil)_dbcAMP differentiated	0.0	93105_Dermal Fibroblasts CCD1070_IL-1 beta 1 ng/ml	15.6
93248_EOL-1 (Eosinophil)_dbcAMP/PMAionomycin	0.0	93772_dermal fibroblast_IFN gamma	24.9
93356_Dendritic Cells_none	0.0	93771_dermal fibroblast_IL-4	34.5
93355_Dendritic Cells_LPS 100 ng/ml	0.0	93892_Dermal fibroblasts_none	5.1
93775_Dendritic Cells_anti-CD40	0.0	99202_Neutrophils_TNFa+LPS	0.0
93774_Monocytes_resting	0.0	99203_Neutrophils_none	0.0
93776_Monocytes_LPS 50 ng/ml	0.0	735010_Colon_normal	1.9
93581_Macrophages_resting	0.0	735019_Lung_none	10.4
93582_Macrophages_LPS 100 ng/ml	0.0	64028-1_Thymus_none	2.8
93098_HUVEC (Endothelial)_none	0.0	64030-1_Kidney_none	10.2
93099_HUVEC (Endothelial)_starved	2.3		

Panel 2.1 Summary: Ag1460 The level of expression of the NOV3 - 24SC113 gene is low in the samples used for Panel 2.1, with highest expression in normal uterus (CT = 32.5). However, this gene appears to be more highly expressed in some samples derived from normal uterus, stomach, kidney and lung when compared to the associated cancer tissue. Thus, based upon its profile, the expression of the 24SC113 gene could be of use as a marker for these normal tissues or as a protein therapeutic for the treatment of gastric, uterine, lung and kidney

cancer. In addition, therapeutic activity of the 24SC113 gene product, through the use of peptides, chimeric molecules or small molecule drugs, may be useful in the therapy of gastric cancer.

Panel 4.1D Summary: Ag1460 Expression of the NOV3 - 24SC113 gene is highest in resting astrocytes (CT = 30.9), suggesting that this gene would be an effective marker for astrocytes. Strikingly, expression of this gene in astrocytes is down regulated after treatment with the inflammatory cytokines TNFa and IL-1. Considering the deleterious effect of these cytokines on astrocytes, we may propose that the protein encoded by the 24SC113 gene may be a trophic factor for astrocytes and thus, that the protein encoded by this gene could be beneficial as a protein therapeutic in the treatment of neurodegenerative diseases associated with inflammation, such as Alzheimer's disease, multiple sclerosis, and stroke. In addition, low but significant expression of the 24SC113 gene is seen in activated and non-activated fibroblasts (dermal and lung).

NOV4 - 24SC128

Expression of gene 24SC128 was assessed using the primer-probe set Ag3976, described in Table 20A. Results from RTQ-PCR runs are shown in Tables 20B and 20C.

Table 20A. Probe Name Ag3976

Primers	Sequences	TM	Length	Start Position	SEQ ID NO
Forward	5'-GCTCTCGAAAGTGGGCTATATT-3'	58.9	22	453	128
Probe	FAM-5'-CACTTTTGTTTTATCTTCTCCAACCACCA-3'-TAMRA	66.9	29	493	129
Reverse	5'-TCTCCTATTCAGGTGACTTTCG-3'	58.5	22	524	130

Table 20B. Panel 2.1

Tissue Name	Relative Expression(%) 2.1x4tm6080f_ag3976_a2	Tissue Name	Relative Expression(%) 2.1x4tm6080f_ag3976_a2
Normal Colon GENPAK 061003	15.0	Kidney Cancer Clontech 9010320	3.5
97759 Colon cancer (OD06064)	8.9	Kidney NAT Clontech 9010321	34.6
97760 Colon cancer NAT (OD06064)	4.0	Kidney Cancer Clontech 8120607	12.7
97778 Colon cancer (OD06159)	4.2	Kidney NAT Clontech 8120608	4.1
97779 Colon cancer NAT (OD06159)	8.1	Normal Uterus GENPAK 061018	47.1
98859 Colon cancer (OD06298-08)	36.8	Uterus Cancer GENPAK 064011	31.1
98860 Colon cancer NAT (OD06298-018)	32.2	Normal Thyroid Clontech A+ 6570-1 (7080817)	2.4

83237 CC Gr 2 ascend colon (OD03921)	19.4	Thyroid Cancer GENPAK 064010	9.5
83238 CC NAT (OD03921)	20.4	Thyroid Cancer INVITROGEN A302152	13 9
97766 Colon cancer metastasis (OD06104)	22.5	Thyroid NAT INVITROGEN A302153	89.5
97767 Lung NAT (OD06104)	37.7	Normal Breast GENPAK 061019	72.8
87472 Colon mets to lung (OD04451-01)	16.6	84877 Breast Cancer (OD04566)	10.0
87473 Lung NAT (OD04451-02)	13.6	Breast Cancer Res. Gen. 1024	27.7
Normal Prostate Clontech A+ 6546-1 (8090438)	9.4	85975 Breast Cancer (OD04590-01)	12.5
84140 Prostate Cancer (OD04410)	3.0	85976 Breast Cancer Mets (OD04590-03)	32.8
84141 Prostate NAT (OD04410)	9.1	87070 Breast Cancer Metastasis (OD04655-05)	95.4
Normal Lung GENPAK 061010	38.5	GENPAK Breast Cancer 064006	4.5
92337 Invasive poor diff. lung adeno (OD04945-01	25.0	Breast Cancer Clontech 9100266	19.4
92338 Lung NAT (OD04945-03)	30.5	Breast NAT Clontech 9100265	35.0
84136 Lung Malignant Cancer (OD03126)	20.6	Breast Cancer INVITROGEN A209073	9.2
84137 Lung NAT (OD03126)	16.6	Breast NAT INVITROGEN A2090734	36.4
90372 Lung Cancer (OD05014A)	14.7	Normal Liver GENPAK 061009	13.3
90373 Lung NAT (OD05014B)	8.2	Liver Cancer Research Genetics RNA 1026	7.9
85950 Lung Cancer (OD04237-01)	18.6	Liver Cancer Research Genetics RNA 1025	23.7
85970 Lung NAT (OD04237-02)	14.9	Paired Liver Cancer Tissue Research Genetics RNA 6004-T	13.6
83255 Ocular Mel Met to Liver (OD04310)	18.8	Paired Liver Tissue Research Genetics RNA 6004-N	8.3
83256 Liver NAT (OD04310)	9.3	Paired Liver Cancer Tissue Research Genetics RNA 6005-T	32.2
84139 Melanoma Mets to Lung (OD04321)	22.5	Paired Liver Tissue Research Genetics RNA 6005-N	8.7
84138 Lung NAT (OD04321)	26.3	Liver Cancer GENPAK 064003	9.4
Normal Kidney GENPAK 061008	23.6	Normal Bladder GENPAK 061001	16.8
83786 Kidney Ca, Nuclear grade 2 (OD04338)	33.0	Bladder Cancer Research Genetics RNA 1023	19.8
83787 Kidney NAT (OD04338)	18.3	Bladder Cancer INVITROGEN A302173	17.5
83788 Kidney Ca Nuclear grade 1/2 (OD04339)	12.6	Normal Ovary Res. Gen.	27.1
83789 Kidney NAT (OD04339)	13.4	Ovarian Cancer GENPAK 064008	4.9
83790 Kidney Ca, Clear cell type (OD04340)	10.0	97773 Ovarian cancer (OD06145)	2.2
83791 Kidney NAT (OD04340)	27.0	97775 Ovarian cancer NAT (OD06145)	37.8
83792 Kidney Ca, Nuclear grade 3 (OD04348)	9.0	Normal Stomach GENPAK 061017	39.1
83793 Kidney NAT (OD04348)	18.6	Gastric Cancer Clontech 9060397	8.4
85973 Kidney Cancer (OD04450-01)	100.0	NAT Stomach Clontech 9060396	8.1

85974 Kidney NAT (OD04450-03)	18.1	Gastric Cancer Clontech 9060395	40.5
Kidney Cancer Clontech 8120613	3.9	NAT Stomach Clontech 9060394	26.5
Kidney NAT Clontech 8120614	6.5	Gastric Cancer GENPAK 064005	18.9

Table 20C. Panel 4.1D

Tissue Name	Relative Expression(%) 4.1dx4tm6081_ag3976_a2	Tissue Name	Relative Expression(%) 4.1dx4tm6081_ag3976_a2
93768_Secondary Th1_anti-CD28/anti-CD3	86.4	93100_HUVEC (Endothelial)_IL-1b	55.1
93769_Secondary Th2_anti-CD28/anti-CD3	70.2	93779_HUVEC (Endothelial)_IFN gamma	67.6
93770_Secondary Tr1_anti-CD28/anti-CD3	57.3	93102_HUVEC (Endothelial)_TNF alpha + IFN gamma	32.8
93573_Secondary Th1_resting day 4-6 in IL-2	16.6	93101_HUVEC (Endothelial)_TNF alpha + IL4	51.5
93572_Secondary Th2_resting day 4-6 in IL-2	28.4	93781_HUVEC (Endothelial)_IL-11	50.1
93571_Secondary Tr1_resting day 4-6 in IL-2	24.6	93583_Lung Microvascular Endothelial Cells_none	94.1
93568_primary Th1_anti-CD28/anti-CD3	58.7	93584_Lung Microvascular Endothelial Cells_TNFa (4 ng/ml) and IL1b (1 ng/ml)	59.7
93569_primary Th2_anti-CD28/anti-CD3	100.0	92662_Microvascular Dermal endothelium_none	14.3
93570_primary Tr1_anti-CD28/anti-CD3	70.4	92663_Microsvasular Dermal endothelium_TNFa (4 ng/ml) and IL1b (1 ng/ml)	24.0
93565_primary Th1_resting dy 4-6 in IL-2	27.5	93773_Bronchial epithelium_TNFa (4 ng/ml) and IL1b (1 ng/ml) **	54.5
93566_primary Th2_resting dy 4-6 in IL-2	13.8	93347_Small Airway Epithelium_none	32.1
93567_primary Tr1_resting dy 4-6 in IL-2	48.9	93348_Small Airway Epithelium_TNFa (4 ng/ml) and IL1b (1 ng/ml)	65.8
93351_CD45RA CD4 lymphocyte_anti-CD28/anti-CD3	57.4	92668_Coronery Artery SMC_resting	32.0
93352_CD45RO CD4 lymphocyte_anti-CD28/anti-CD3	68.7	92669_Coronery Artery SMC_TNFa (4 ng/ml) and IL1b (1 ng/ml)	25.5
93251_CD8 Lymphocytes_anti-CD28/anti-CD3	91.9	93107_astrocytes_resting	30.9
93353_chronic CD8 Lymphocytes 2ry_resting dy 4-6 in IL-2	76.2	93108_astrocytes_TNFa (4 ng/ml) and IL1b (1 ng/ml)	18.4
93574_chronic CD8 Lymphocytes 2ry_activated CD3/CD28	42.0	92666_KU-812 (Basophil)_resting	32.3
93354_CD4_none	25.1	92667_KU-812 (Basophil)_PMA/ionoycin	36.0
93252_Secondary Th1/Th2/Tr1_anti-CD95 CH11	37.9	93579_CCD1106 (Keratinocytes)_none	82.5
93103_LAK cells_resting	45.0	93580_CCD1106 (Keratinocytes)_TNFa and	56.6

		IFNg **	
93788_LAK cells_IL-2	58.1	93791_Liver Cirrhosis	7.8
93787_LAK cells_IL-2+IL-12	48.5	93577_NCI-H292	31.2
93789_LAK cells_IL-2+IFN gamma	44.6	93358_NCI-H292_IL-4	53.9
93790_LAK cells_IL-2+ IL-18	70.0	93360_NCI-H292_IL-9	56.7
93104_LAK cells_PMA/ionomycin and IL-18	19.9	93359_NCI-H292_IL-13	52.6
93578_NK Cells IL-2_resting	51.3	93357_NCI-H292_IFN gamma	38.8
93109_Mixed Lymphocyte Reaction_Two Way MLR	68.7	93777_HPAEC -	23.4
93110_Mixed Lymphocyte Reaction_Two Way MLR	57.2	93778_HPAEC_IL-1 beta/TNA alpha	28.2
93111_Mixed Lymphocyte Reaction_Two Way MLR	49.9	93254_Normal Human Lung Fibroblast_none	27.7
93112_Mononuclear Cells (PBMCS)_resting	16.1	93253_Normal Human Lung Fibroblast_TNFa (4 ng/ml) and IL-1b (1 ng/ml)	17.2
93113_Mononuclear Cells (PBMCS)_PWM	49.7	93257_Normal Human Lung Fibroblast_IL-4	32.8
93114_Mononuclear Cells (PBMCS)_PHA-L	57.0	93256_Normal Human Lung Fibroblast_IL-9	52.1
93249_Ramos (B cell)_none	59.6	93255_Normal Human Lung Fibroblast_IL-13	49.4
93250_Ramos (B cell)_ionomycin	79.4	93258_Normal Human Lung Fibroblast_IFN gamma	40.9
93349_B lymphocytes_PWM	67.0	93106_Dermal Fibroblasts CCD1070_resting	49.9
93350_B lymphoytes_CD40L and IL-4	64.2	93361_Dermal Fibroblasts CCD1070_TNF alpha 4 ng/ml	61.9
92665_EOL-1 (Eosinophil)_dbcAMP differentiated	66.9	93105_Dermal Fibroblasts CCD1070_IL-1 beta 1 ng/ml	31.4
93248_EOL-1 (Eosinophil)_dbcAMP/PMAion omycin	43.9	93772_dermal fibroblast_IFN gamma	23.0
93356_Dendritic Cells_none	26.0	93771_dermal fibroblast_IL-4	30.4
93355_Dendritic Cells_LPS 100 ng/ml	20.9	93892_Dermal fibroblasts_none	19.0
93775_Dendritic Cells_anti-CD40	30.1	99202_Neutrophils_TNFa+LPS	1.5
93774_Monocytes_resting	16.0	99203_Neutrophils_none	1.0
93776_Monocytes_LPS 50 ng/ml	11.7	735010_Colon_normal	12.3
93581_Macrophages_resting	33.9	735019_Lung_none	17.7
93582_Macrophages_LPS 100 ng/ml	10.9	64028-1_Thymus_none	35.8
93098_HUVEC (Endothelial)_none	37.4	64030-1_Kidney_none	30.3
93099_HUVEC (Endothelial)_starved	38.3		

Panel 2.1 Summary: Ag3976 The NOV4 - 24SC128 gene is fairly ubiquitously expressed at moderate levels in the cancer tissues as well as the normal adjacent tissues used for Panel 2.1. However, a high level of expression is seen in a kidney cancer sample (CT = 29.7) when compared to its associated normal adjacent tissue (CT = 32.2), as well as in a single sample

of metastatic breast cancer (CT = 29.7). Thus, based upon this profile, expression of the 24SC128 gene could be of use as a marker for a form of renal or breast cancer. In addition, therapeutic inhibition of the activity of this gene product, through the use of antibodies or small molecule drugs, may be useful in the treatment of renal or breast cancer.

Panel 4.1D Summary: Ag3976 The NOV4 - 24SC128 gene is ubiquitously expressed at a moderate levels in activated T cells (CD4 and CD8), B cells, eosinophils, endothelial cells (HUVEC and lung microvasculature endothelial cells) and fibroblasts. Interestingly, 24SC128 gene expression appears to be up-regulated in TH1 and TH2 cells upon activation, suggesting a role for this gene in T cell-mediated diseases such as asthma, delayed type hypersensitivity, infectious disease, and autoimmune disease (rheumatoid arthritis, inflammatory bowel disease, and psoriasis).

NOV8 - 24SC714

Expression of gene 24SC714 was assessed using the primer-probe set Ag4002, described in Table 21A. Results from RTQ-PCR runs are shown in Tables 21B and 21C.

Table 21A. Probe Name Ag4002

Primers	Sequences	TM	Length	Start Position	SEQ ID NO
Forward	5'-GCCCTGATCAAGTTTTCATACC-3'	59.8	22	364	131
Probe	FAM-5'-CACATAGCTCAGCCTGCTCTGAGTTGA-3'-TAMRA	69	27	387	132
Reverse	5'-TGTCAACTCCACATGAATCAAA-3'	59	22	428	133

Table 21B. Panel 2.1

Tissue Name	Relative Expression(%) 2.1x4tm6143f_ag4002_b1	Tissue Name	Relative Expression(%) 2.1x4tm6143f_ag4002_b1
Normal Colon GENPAK 061003	2.8	Kidney Cancer Clontech 9010320	0.0
97759 Colon cancer (OD06064)	0.0	Kidney NAT Clontech 9010321	7.5
97760 Colon cancer NAT (OD06064)	0.0	Kidney Cancer Clontech 8120607	0.0
97778 Colon cancer (OD06159)	0.0	Kidney NAT Clontech 8120608	0.0
97779 Colon cancer NAT (OD06159)	0.0	Normal Uterus GENPAK 061018	2.7
98859 Colon cancer (OD06298-08)	28.5	Uterus Cancer GENPAK 064011	0.0
98860 Colon cancer NAT (OD06298-018)	2.1	Normal Thyroid Clontech A+ 6570-1 (7080817)	0.0
83237 CC Gr.2 ascend colon (OD03921)	5.1	Thyroid Cancer GENPAK 064010	0.0
83238 CC NAT (OD03921)	3.6	Thyroid Cancer INVITROGEN A302152	0.0

97766 Colon cancer metastasis (OD06104)	20.4	Thyroid NAT INVITROGEN A302153	0.0
97767 Lung NAT (OD06104)	2.6	Normal Breast GENPAK 061019	5.4
87472 Colon mets to lung (OD04451-01)	76.7	84877 Breast Cancer (OD04566)	0.0
87473 Lung NAT (OD04451-02)	1.3	Breast Cancer Res. Gen. 1024	2.5
Normal Prostate Clontech A+ 6546-1 (8090438)	0.0	85975 Breast Cancer (OD04590-01)	0.0
84140 Prostate Cancer (OD04410)	0.0	85976 Breast Cancer Mets (OD04590-03)	10.5
84141 Prostate NAT (OD04410)	0.0	87070 Breast Cancer Metastasis (OD04655-05)	3.0
Normal Lung GENPAK 061010	16.2	GENPAK Breast Cancer 064006	0.0
92337 Invasive poor diff. lung adeno (OD04945-01	24.4	Breast Cancer Clontech 9100266	0.0
92338 Lung NAT (OD04945-03)	32.7	Breast NAT Clontech 9100265	0.0
84136 Lung Malignant Cancer (OD03126)	3.4	Breast Cancer INVITROGEN A209073	0.0
84137 Lung NAT (OD03126)	12.6	Breast NAT INVITROGEN A2090734	3.3
90372 Lung Cancer (OD05014A)	8.2	Normal Liver GENPAK 061009	0.0
90373 Lung NAT (OD05014B)	100.0	Liver Cancer Research Genetics RNA 1026	0.0
85950 Lung Cancer (OD04237-01)	5.4	Liver Cancer Research Genetics RNA 1025	0.0
85970 Lung NAT (OD04237-02)	32.8	Paired Liver Cancer Tissue Research Genetics RNA 6004-T	0.0
83255 Ocular Mel Met to Liver (OD04310)	3.3	Paired Liver Tissue Research Genetics RNA 6004-N	0.0
83256 Liver NAT (OD04310)	0.0	Paired Liver Cancer Tissue Research Genetics RNA 6005-T	0.0
84139 Melanoma Mets to Lung (OD04321)	0.0	Paired Liver Tissue Research Genetics RNA 6005-N	0.0
84138 Lung NAT (OD04321)	0.0	Liver Cancer GENPAK 064003	0.0
Normal Kidney GENPAK 061008	6.1	Normal Bladder GENPAK 061001	0.0
83786 Kidney Ca, Nuclear grade 2 (OD04338)	0.0	Bladder Cancer Research Genetics RNA 1023	6.9
83787 Kidney NAT (OD04338)	2.6	Bladder Cancer INVITROGEN A302173	9.9
83788 Kidney Ca Nuclear grade 1/2 (OD04339)	0.0	Normal Ovary Res. Gen.	0.0
83789 Kidney NAT (OD04339)	0.0	Ovarian Cancer GENPAK 064008	0.0
83790 Kidney Ca, Clear cell type (OD04340)	0.0	97773 Ovarian cancer (OD06145)	0.0
83791 Kidney NAT (OD04340)	1.5	97775 Ovarian cancer NAT (OD06145)	0.0
83792 Kidney Ca, Nuclear grade 3 (OD04348)	2.8	Normal Stomach GENPAK 061017	5.0
83793 Kidney NAT (OD04348)	0.0	Gastric Cancer Clontech 9060397	9.9
85973 Kidney Cancer (OD04450-01)	0.0	NAT Stomach Clontech 9060396	2.2
85974 Kidney NAT (OD04450-03)	0.0	Gastric Cancer Clontech 9060395	0.0
Kidney Cancer Clontech 8120613	0.0	NAT Stomach Clontech 9060394	0.0

Kidney NAT Clontech 8120614	0.0	Gastric Cancer GENPAK 064005	0.0

Table 21C. Panel 4.1D

Tissue Name	Relative Expression(%) 4.1dtm6147f_ag4002	Tissue Name	Relative Expression(%) 4.1dtm6147f_ag4002
93768_Secondary Th1_anti-CD28/anti-CD3	0.0	93100_HUVEC (Endothelial)_IL-1b	3.9
93769_Secondary Th2_anti-CD28/anti-CD3	0.0	93779_HUVEC (Endothelial)_IFN gamma	15.7
93770_Secondary Tr1_anti-CD28/anti-CD3	0.0	93102_HUVEC (Endothelial)_TNF alpha + IFN gamma	20.6
93573_Secondary Th1_resting day 4-6 in IL-2	0.0	93101_HUVEC (Endothelial)_TNF alpha + IL4	26.1
93572_Secondary Th2_resting day 4-6 in IL-2	0.0	93781_HUVEC (Endothelial)_IL-11	0.0
93571_Secondary Tr1_resting day 4-6 in IL-2	0.0	93583_Lung Microvascular Endothelial Cells_none	3.7
93568_primary Th1_anti-CD28/anti-CD3	0.0	93584_Lung Microvascular Endothelial Cells_TNFa (4 ng/ml) and IL1b (1 ng/ml)	2.4
93569_primary Th2_anti-CD28/anti-CD3	0.0	92662_Microvascular Dermal endothelium_none	5.3
93570_primary Tr1_anti-CD28/anti-CD3	0.0	92663_Microsvasular Dermal endothelium_TNFa (4 ng/ml) and IL1b (1 ng/ml)	1.9
93565_primary Th1_resting dy 4-6 in IL-2	0.0	93773_Bronchial epithelium_TNFa (4 ng/ml) and IL1b (1 ng/ml) **	0.0
93566_primary Th2_resting dy 4-6 in IL-2	0.0	93347_Small Airway Epithelium_none	1.2
93567_primary Tr1_resting dy 4-6 in IL-2	0.0	93348_Small Airway Epithelium_TNFa (4 ng/ml) and IL1b (1 ng/ml)	0.0
93351_CD45RA CD4 lymphocyte_anti-CD28/anti-CD3	0.0	92668_Coronery Artery SMC_resting	0.0
93352_CD45RO CD4 lymphocyte_anti-CD28/anti-CD3	0.0	92669_Coronery Artery SMC_TNFa (4 ng/ml) and IL1b (1 ng/ml)	3.1
93251_CD8 Lymphocytes_anti-CD28/anti-CD3	0.0	93107_astrocytes_resting	1.0
93353_chronic CD8 Lymphocytes 2ry_resting dy 4-6 in IL-2	0.0	93108_astrocytes_TNFa (4 ng/ml) and IL1b (1 ng/ml)	0.0
93574_chronic CD8 Lymphocytes 2ry_activated CD3/CD28	0.0	92666_KU-812 (Basophil)_resting	0.0
93354_CD4_none	0.0	92667_KU-812 (Basophil)_PMA/ionoycin	0.0
93252_Secondary Th1/Th2/Tr1_anti-CD95 CH11	1.0	93579_CCD1106 (Keratinocytes)_none	0.0
93103_LAK cells_resting	0.0	93580_CCD1106 (Keratinocytes)_TNFa and IFNg **	0.0
93788_LAK cells_IL-2	0.0	93791_Liver Cirrhosis	0.0
93787_LAK cells_IL-2+IL-12	0.0	93577_NCI-H292	0.0

93789_LAK cells_IL-2+IFN gamma	0.0	93358_NCI-H292_IL-4	0.0
93790_LAK cells_IL-2+ IL-18	0.0	93360_NCI-H292_IL-9	0.0
93104_LAK cells_PMA/ionomycin and IL-18	0.0	93359_NCI-H292_IL-13	0.0
93578_NK Cells IL-2_resting	0.0	93357_NCI-H292_IFN gamma	0.0
93109_Mixed Lymphocyte Reaction_Two Way MLR	1.7	93777_HPAEC_-	1.4
93110_Mixed Lymphocyte Reaction_Two Way MLR	0.0	93778_HPAEC_IL-1 beta/TNA alpha	0.0
93111_Mixed Lymphocyte Reaction_Two Way MLR	0.0	93254_Normal Human Lung Fibroblast_none	0.0
93112_Mononuclear Cells (PBMCs)_resting	0.9	93253_Normal Human Lung Fibroblast_TNFa (4 ng/ml) and IL-1b (1 ng/ml)	0.0
93113_Mononuclear Cells (PBMCs)_PWM	0.0	93257_Normal Human Lung Fibroblast_IL-4	0.9
93114_Mononuclear Cells (PBMCs)_PHA-L	0.0	93256_Normal Human Lung Fibroblast_IL-9	0.9
93249_Ramos (B cell)_none	0.0	93255_Normal Human Lung Fibroblast_IL-13	4.0
93250_Ramos (B cell)_ionomycin	0.0	93258_Normal Human Lung Fibroblast_IFN gamma	2.5
93349_B lymphocytes_PWM	0.0	93106_Dermal Fibroblasts CCD1070_resting	6.8
93350_B lymphoytes_CD40L and IL-4	1.5	93361_Dermal Fibroblasts CCD1070_TNF alpha 4 ng/ml	6.9
92665_EOL-1 (Eosinophil)_dbcAMP differentiated	0.0	93105_Dermal Fibroblasts CCD1070_IL-1 beta 1 ng/ml	0.0
93248_EOL-1 (Eosinophil)_dbcAMP/PMAion omycin	0.0	93772_dermal fibroblast_IFN gamma	0.0
93356_Dendritic Cells_none	1.0	93771_dermal fibroblast_IL-4	0.0
93355_Dendritic Cells_LPS 100 ng/ml	0.0	93892_Dermal fibroblasts_none	0.8
93775_Dendritic Cells_anti-CD40	0.0	99202_Neutrophils_TNFa+LPS	0.9
93774_Monocytes_resting	0.0	99203_Neutrophils_none	0.0
93776_Monocytes_LPS 50 ng/ml	0.0	735010_Colon_normal	6.7
93581_Macrophages_resting	2.1	735019_Lung_none	12.5
93582_Macrophages_LPS 100 ng/ml	0.0	64028-1_Thymus_none	32.8
93098_HUVEC (Endothelial)_none	7.8	64030-1_Kidney_none	100.0
93099_HUVEC (Endothelial)_starved	11.0		

Panel 2.1 Summary: Ag4002 The NOV8 - 24SC714 gene is expressed at low levels in normal lung tissues but to a lesser degree in the associated lung tumor tissues in Panel 2.1. Low but significant expression of this gene is also seen in a metastatic colon cancer sample (CT = 33.8) when compared to its associated normal adjacent tissue. Thus, based upon this profile, the expression of the 24SC714 gene could be of use as a marker for normal lung or colon cancer. In addition, therapeutic inhibition of the activity of this gene product, through the use of antibodies

or small molecule drugs, may be useful in the treatment of colon cancer. Furthermore, peptides, chimeric molecules and small molecule drugs might be useful in the therapy of lung cancer.

Panel 4.1D Summary: Ag4002 Expression of this NOV8 gene is highest in normal kidney (CT = 31.2). In addition, the NOV8 - 24SC714 gene is expressed at low levels in HUVECs independent of treatment with cytokines (CT values = 33 to 35). Consistent with these data, this gene is also expressed in endothelial cells from lung and dermis, independent of activation status. Therefore, antibody or protein therapeutic against the protein encoded by the 24SC714 gene could be useful in the treatment of inflammation.

NOV10a - 100340173

Expression of gene 100340173 (NOV10a) was assessed using the primer-probe set Ag4001, described in Table 22A. Results from RTQ-PCR runs are shown in Tables 22B and 22C.

Table 22A. Probe Name Ag4001

Primers	Sequences	TM	Length	Start Position	SEQ ID NO
Forward	5'-TCCTACCCAGCTTCTGAATTCT-3'	59.4	22	633	134
Probe	FAM-5'-TACTTGGGTACCACCCTGCGGACAAT-3'-TAMRA	70.8	26	655	135
Reverse	5'-AACACTCTGTTCTGCAATGACA-3'	58.4	22	687	136

Table 22B. Panel 2.1

Tissue Name	Relative Expression(%) 2.1dx4tm6143f_ag4001_a2	Tissue Name	Relative Expression(%) 2.1dx4tm6143f_ag4001_a2
Normal Colon GENPAK 061003	19.3	Kidney Cancer Clontecn 9010320	14.8
97759 Colon cancer (OD06064)	30.9	Kidney NAT Clontech 9010321	79.9
97760 Colon cancer NAT (OD06064)	10.4	Kidney Cancer Clontech 8120607	28.2
97778 Colon cancer (OD06159)	11.3	Kidney NAT Clontech 8120608	8.6
97779 Colon cancer NAT (OD06159)	8.6	Normal Uterus GENPAK 061018	78.1
98859 Colon cancer (OD06298-08)	32.3	Uterus Cancer GENPAK 064011	10.6
98860 Colon cancer NAT (OD06298-018)	17.4	Normal Thyroid Clontech A+ 6570-1 (7080817)	5.0
83237 CC Gr.2 ascend colon (OD03921)	13.3	Thyroid Cancer GENPAK 064010	28.4
83238 CC NAT (OD03921)	14.1	Thyroid Cancer INVITROGEN A302152	7.7
97766 Colon cancer metastasis (OD06104)	8.7	Thyroid NAT INVITROGEN A302153	31.8
97767 Lung NAT (OD06104)	96.0	Normal Breast GENPAK 061019	31.0

87472 Colon mets to lung (OD04451-01)	9.7	84877 Breast Cancer (OD04566)	1.4
87473 Lung NAT (OD04451-02)	42.3	Breast Cancer Res. Gen. 1024	11.2
Normal Prostate Clontech A+ 6546-1 (8090438)	4.5	85975 Breast Cancer (OD04590-01)	3.7
84140 Prostate Cancer (OD04410)	3.8	85976 Breast Cancer Mets (OD04590-03)	0.0
84141 Prostate NAT (OD04410)	26.6	87070 Breast Cancer Metastasis (OD04655-05)	13.7
Normal Lung GENPAK 061010	38.0	GENPAK Breast Cancer 064006	1.9
92337 Invasive poor diff. lung adeno (OD04945-01	6.9	Breast Cancer Clontech 9100266	6.5
92338 Lung NAT (OD04945-03)	47.4	Breast NAT Clontech 9100265	25.4
84136 Lung Malignant Cancer (OD03126)	7.9	Breast Cancer INVITROGEN A209073	16.8
84137 Lung NAT (OD03126)	24.4	Breast NAT INVITROGEN A2090734	28.2
90372 Lung Cancer (OD05014A)	17.8	Normal Liver GENPAK 061009	40.8
90373 Lung NAT (OD05014B)	45.2	Liver Cancer Research Genetics RNA 1026	6.1
85950 Lung Cancer (OD04237-01)	3.3	Liver Cancer Research Genetics RNA 1025	32.8
85970 Lung NAT (OD04237-02)	26.6	Paired Liver Cancer Tissue Research Genetics RNA 6004-T	22.1
83255 Ocular Mel Met to Liver (OD04310)	20.7	Paired Liver Tissue Research Genetics RNA 6004-N	7.9
83256 Liver NAT (OD04310)	16.0	Paired Liver Cancer Tissue Research Genetics RNA 6005-T	27.8
84139 Melanoma Mets to Lung (OD04321)	43.8	Paired Liver Tissue Research Genetics RNA 6005-N	28.1
84138 Lung NAT (OD04321)	46.2	Liver Cancer GENPAK 064003	12.6
Normal Kidney GENPAK 061008	21.6	Normal Bladder GENPAK 061001	12.8
83786 Kidney Ca, Nuclear grade 2 (OD04338)	36.7	Bladder Cancer Research Genetics RNA 1023	3.1
83787 Kidney NAT (OD04338)	35.1	Bladder Cancer INVITROGEN A302173	12.8
83788 Kidney Ca Nuclear grade 1/2 (OD04339)	14.3	Normal Ovary Res. Gen.	8.9
83789 Kidney NAT (OD04339)	13.0	Ovarian Cancer GENPAK 064008	6.8
83790 Kidney Ca, Clear cell type (OD04340)	21.5	97773 Ovarian cancer (OD06145)	3.2
83791 Kidney NAT (OD04340)	23.2	97775 Ovarian cancer NAT (OD06145)	18.8
83792 Kidney Ca, Nuclear grade 3 (OD04348)	7.2	Normal Stomach GENPAK 061017	22.4
83793 Kidney NAT (OD04348)	19.5	Gastric Cancer Clontech 9060397	9.1
85973 Kidney Cancer (OD04450-01)	100.0	NAT Stomach Clontech 9060396	6.8
85974 Kidney NAT (OD04450-03)	29.7	Gastric Cancer Clontech 9060395	24.3
Kidney Cancer Clontech 8120613	0.5	NAT Stomach Clontech 9060394	17.8
Kidney NAT Clontech 8120614	7.0	Gastric Cancer GENPAK 064005	9.9

Table 22C. Panel 4.1D

Tissue Name	Relative Expression(%) 4.1dtm6146f_ag4001	Tissue Name	Relative Expression(%) 4.1dtm6146f_ag4001
93768_Secondary Th1_anti-CD28/anti-CD3	20.0	93100_HUVEC (Endothelial)_IL-1b	20.3
93769_Secondary Th2_anti-CD28/anti-CD3	28.5	93779_HUVEC (Endothelial)_IFN gamma	25.5
93770_Secondary Tr1_anti-CD28/anti-CD3	23.7	93102_HUVEC (Endothelial)_TNF alpha + IFN gamma	9.2
93573_Secondary Th1_resting day 4-6 in IL-2	4.6	93101_HUVEC (Endothelial)_TNF alpha + IL4	11.9
93572_Secondary Th2_resting day 4-6 in IL-2	5.5	93781_HUVEC (Endothelial)_IL-11	8.6
93571_Secondary Tr1_resting day 4-6 in IL-2	4.3	93583_Lung Microvascular Endothelial Cells_none	32.1
93568_primary Th1_anti-CD28/anti-CD3	15.9	93584_Lung Microvascular Endothelial Cells_TNFa (4 ng/ml) and IL1b (1 ng/ml)	14.8
93569_primary Th2_anti-CD28/anti-CD3	18.4	92662_Microvascular Dermal endothelium_none	17.1
93570_primary Tr1_anti-CD28/anti-CD3	20.2	92663_Microsvasular Dermal endothelium_TNFa (4 ng/ml) and IL1b (1 ng/ml)	10.5
93565_primary Th1_resting dy 4-6 in IL-2	3.4	93773_Bronchial epithelium_TNFa (4 ng/ml) and IL1b (1 ng/ml) **	14.4
93566_primary Th2_resting dy 4-6 in IL-2	1.8	93347_Small Airway Epithelium_none	6.9
93567_primary Tr1_resting dy 4-6 in IL-2	3.2	93348_Small Airway Epithelium_TNFa (4 ng/ml) and IL1b (1 ng/ml)	14.3
93351_CD45RA CD4 lymphocyte_anti-CD28/anti-CD3	18.2	92668_Coronery Artery SMC_resting	15.2
93352_CD45RO CD4 lymphocyte_anti-CD28/anti-CD3	28.9	92669_Coronery Artery SMC_TNFa (4 ng/ml) and IL1b (1 ng/ml)	14.5
93251_CD8 Lymphocytes_anti-CD28/anti-CD3	30.1	93107_astrocytes_resting	24.1
93353_chronic CD8 Lymphocytes 2ry_resting dy 4-6 in IL-2	20.4	93108_astrocytes_TNFa (4 ng/ml) and IL1b (1 ng/ml)	15.0
93574_chronic CD8 Lymphocytes 2ry_activated CD3/CD28	10.6	92666_KU-812 (Basophil)_resting	42.3
93354_CD4_none	3.8	92667_KU-812 (Basophil)_PMA/ionoycin	100.0
93252_Secondary Th1/Th2/Tr1_anti-CD95 CH11	4.0	93579_CCD1106 (Keratinocytes)_none	17.1
93103_LAK cells_resting	12.7	93580_CCD1106 (Keratinocytes)_TNFa and IFNg **	10.9
93788_LAK cells_IL-2	21.0	93791_Liver Cirrhosis	7.1
93787_LAK cells_IL-2+IL-12	15.0	93577_NCI-H292	6.7
93789_LAK cells_IL-2+IFN gamma	16.4	93358_NCI-H292_IL-4	10.2
93790_LAK cells_IL-2+ IL-18	19.8	93360_NCI-H292_IL-9	16.5

93104_LAK cells_PMA/ionomycin and IL-18	22.5	93359_NCI-H292_IL-13	20.7
93578_NK Cells IL-2_resting	42.9	93357_NCI-H292_IFN gamma	12.9
93109_Mixed Lymphocyte Reaction_Two Way MLR	20.7	93777_HPAEC_-	14.9
93110_Mixed Lymphocyte Reaction_Two Way MLR	16.0	93778_HPAEC_IL-1 beta/TNA alpha	16.8
93111_Mixed Lymphocyte Reaction_Two Way MLR	16.3	93254_Normal Human Lung Fibroblast_none	16.3
93112_Mononuclear Cells (PBMCs)_resting	4.0	93253_Normal Human Lung Fibroblast_TNFa (4 ng/ml) and IL-1b (1 ng/ml)	8.2
93113_Mononuclear Cells (PBMCs)_PWM	15.8	93257_Normal Human Lung Fibroblast_IL-4	21.8
93114_Mononuclear Cells (PBMCs)_PHA-L	12.3	93256_Normal Human Lung Fibroblast_IL-9	22.7
93249_Ramos (B cell)_none	24.8	93255_Normal Human Lung Fibroblast_IL-13	20.2
93250_Ramos (B cell)_ionomycin	28.3	93258_Normal Human Lung Fibroblast_IFN gamma	22.2
93349_B lymphocytes_PWM	16.6	93106_Dermal Fibroblasts CCD1070_resting	14.6
93350_B lymphoytes_CD40L and IL-4	11.9	93361_Dermal Fibroblasts CCD1070_TNF alpha 4 ng/ml	17.6
92665_EOL-1 (Eosinophil)_dbcAMP differentiated	10.2	93105_Dermal Fibroblasts CCD1070_IL-1 beta 1 ng/ml	13.9
93248_EOL-1 (Eosinophil)_dbcAMP/PMAion omycin	4.1	93772_dermal fibroblast_IFN gamma	11.9
93356_Dendritic Cells_none	9.7	93771_dermal fibroblast_IL-4	17.6
93355_Dendritic Cells_LPS 100 ng/ml	5.6	93892_Dermal fibroblasts_none	7.4
93775_Dendritic Cells_anti-CD40	11.4	99202_Neutrophils_TNFa+LPS	1.4
93774_Monocytes_resting	3.8	99203_Neutrophils_none	0.8
93776_Monocytes_LPS 50 ng/ml	7.4	735010_Colon_normal	7.9
93581_Macrophages_resting	7.3	735019_Lung_none	17.7
93582_Macrophages_LPS 100 ng/ml	4.5	64028-1_Thymus_none	12.9
93098_HUVEC (Endothelial)_none	9.2	64030-1_Kidney_none	18.8
93099_HUVEC (Endothelial)_starved	13.7		

Panel 2.1 Summary: Ag4001 The NOV10 - 100340173 gene is expressed at low to moderate levels across the majority of samples on this panel, with highest expression detected in a kidney cancer sample (CT = 29.2). Thus, this gene is likely to be involved in proliferation and survival of many different cell types. Specific therapeutic inhibition of the activity of this gene product, through the use of antibodies or small molecule drugs, may therefore be useful in the treatment of many different forms of cancer.

Panel 4.1D Summary: Ag4001 The NOV10 - 100340173 gene is ubiquitously expressed at low to moderate levels in the majority of samples on this panel (CT values = 30-33). Interestingly, this gene is highly expressed in basophils after activation by treatment with

PMA/ionomycin (CT = 27.4). Therefore, the protein encoded for by the 100340173 gene could play a role in the development of allergies. Antibodies against this protein could thus be used to reduce or inhibit inflammation observed in allergy, asthma, and psoriasis. In addition, 100340173 gene expression is up-regulated in activated TH1 and TH2 cells, further suggesting that modulation of the protein encoded by this gene might be important in immune-mediated disease.

NOV12 - 87917235

Expression of gene 87917235 was assessed using the primer-probe set Ag4003, described in Table 23A.

Table 23A. Probe Name Ag4003

Primers	Sequences	TM	Length	Start Position	SEQ ID NO
Forward	5'-ATATGATTGAGAAGGCCCAAAC-3'	59.3	22	765	137
Probe	FAM-5'-CCTTTAAATTTAGATCTGTGTCTCCCCA-3'-TAMRA	65.3	29	789	138
Reverse	5'-CTGTGTCTCCAGAGAGGTCTGA-3'	59.6	22	818	139

Expression of this NOV12 gene is low/undetectable (CT values > 35) across all of the samples on Panel 4.1D (data not shown).

NOV13 - 87919652

Expression of gene NOV13 - 87919652 was assessed using the primer-probe set Ag4004, described in Table 24A. Results from RTQ-PCR runs are shown in Tables 24B and 24C.

Table 24A. Probe Name Ag4004

Primers	Sequences	TM	Length	Start Position	SEQ ID NO
Forward	5'-CTGGACAGGTTAGGGCTTTG-3'	59.7	20	883	140
Probe	FAM-5'-CCTTCTGGAAGTCTGCCAGTGTCCTT-3'-TAMRA	68.9	26	908	141
Reverse	5'-TGAGAGAGTTCTGGGTGTCCTA-3'	58.9	22	939	142

Table 24B. Panel 2.1

Tissue Name	Relative Expression(%) 2.1dx4tm6143f_ag4004_b2	Tissue Name	Relative Expression(%) 2.1dx4tm6143f_ag4004_b2
Normal Colon GENPAK 061003	1.1	Kidney Cancer Clontech 9010320	1.6
97759 Colon cancer (OD06064)	1.6	Kidney NAT Clontech 9010321	6.1
97760 Colon cancer NAT (OD06064)	2.1	Kidney Cancer Clontech 8120607	1.6

97778 Colon cancer (OD06159)	1.5	Kidney NAT Clontech 8120608	0.2
97779 Colon cancer NAT (OD06159)	2.0	Normal Uterus GENPAK 061018	1.7
98859 Colon cancer (OD06298-08)	5.9	Uterus Cancer GENPAK 064011	1.1
98860 Colon cancer NAT (OD06298-018)	3.5	Normal Thyroid Clontech A+ 6570-1 (7080817)	0.0
83237 CC Gr.2 ascend colon (OD03921)	0.6	Thyroid Cancer GENPAK 064010	1.4
83238 CC NAT (OD03921)	2.0	Thyroid Cancer INVITROGEN A302152	0.6
97766 Colon cancer metastasis (OD06104)	2.1	Thyroid NAT INVITROGEN A302153	1.5
97767 Lung NAT (OD06104)	100.0	Normal Breast GENPAK 061019	2.8
87472 Colon mets to lung (OD04451-01)	1.0	84877 Breast Cancer (OD04566)	0.7
87473 Lung NAT (OD04451-02)	4.4	Breast Cancer Res. Gen. 1024	2.1
Normal Prostate Clontech A+ 6546-1 (8090438)	0.6	85975 Breast Cancer (OD04590-01)	0.6
84140 Prostate Cancer (OD04410)	0.5	85976 Breast Cancer Mets (OD04590-03)	3.4
84141 Prostate NAT (OD04410)	1.1	87070 Breast Cancer Metastasis (OD04655-05)	4.2
Normal Lung GENPAK 061010	14.5	GENPAK Breast Cancer 064006	1.0
92337 Invasive poor diff. lung adeno (OD04945-01	3.4	Breast Cancer Clontech 9100266	0.5
92338 Lung NAT (OD04945-03)	10.9	Breast NAT Clontech 9100265	0.9
84136 Lung Malignant Cancer (OD03126)	1.1	Breast Cancer INVITROGEN A209073	1.1
84137 Lung NAT (OD03126)	2.7	Breast NAT INVITROGEN A2090734	2.4
90372 Lung Cancer (OD05014A)	3.9	Normal Liver GENPAK 061009	2.5
90373 Lung NAT (OD05014B)	1.5	Liver Cancer Research Genetics RNA 1026	0.5
85950 Lung Cancer (OD04237-01)	3.1	Liver Cancer Research Genetics RNA 1025	3.6
85970 Lung NAT (OD04237-02)	5.9	Paired Liver Cancer Tissue Research Genetics RNA 6004-T	2.4
83255 Ocular Mel Met to Liver (OD04310)	0.0	Paired Liver Tissue Research Genetics RNA 6004-N	1.6
83256 Liver NAT (OD04310)	1.4	Paired Liver Cancer Tissue Research Genetics RNA 6005-T	2.7
84139 Melanoma Mets to Lung (OD04321)	1.1	Paired Liver Tissue Research Genetics RNA 6005-N	2.8
84138 Lung NAT (OD04321)	3.9	Liver Cancer GENPAK 064003	2.0
Normal Kidney GENPAK 061008	0.5	Normal Bladder GENPAK 061001	2.1
83786 Kidney Ca, Nuclear grade 2 (OD04338)	2.8	Bladder Cancer Research Genetics RNA 1023	1.8
83787 Kidney NAT (OD04338)	0.7	Bladder Cancer INVITROGEN A302173	3.5
83788 Kidney Ca Nuclear grade 1/2 (OD04339)	1.1	Normal Ovary Res. Gen.	1.4
83789 Kidney NAT (OD04339)	0.3	Ovarian Cancer GENPAK 064008	0.5
83790 Kidney Ca, Clear cell type (OD04340)	0.7	97773 Ovarian cancer (OD06145)	0.0

83791 Kidney NAT (OD04340)	0.8	97775 Ovarian cancer NAT (OD06145)	0 9
83792 Kidney Ca, Nuclear grade 3 (OD04348)	1.2	Normal Stomach GENPAK 061017	13.1
83793 Kidney NAT (OD04348)	1.9	Gastric Cancer Clontech 9060397	0.9
85973 Kidney Cancer (OD04450-01)	0.7	NAT Stomach Clontech 9060396	6.5
85974 Kidney NAT (OD04450-03)	0.2	Gastric Cancer Clontech 9060395	7.8
Kidney Cancer Clontech 8120613	0.2	NAT Stomach Clontech 9060394	8 3
Kidney NAT Clontech 8120614	0.0	Gastric Cancer GENPAK 064005	4.6

Table 24C. Panel 4.1D

Tissue Name	Relative Expression(%) 4.1dtm6148f_ag4004	Tissue Name	Relative Expression(%) 4.1dtm6148f_ag4004
93768_Secondary Th1_anti-CD28/anti-CD3	52.5	93100_HUVEC (Endothelial)_IL-1b	0.0
93769_Secondary Th2_anti-CD28/anti-CD3	55.5	93779_HUVEC (Endothelial)_IFN gamma	0 0
93770_Secondary Tr1_anti-CD28/anti-CD3	44.4	93102_HUVEC (Endothelial)_TNF alpha + IFN gamma	0.0
93573_Secondary Th1_resting day 4-6 in IL-2	31.9	93101_HUVEC (Endothelial)_TNF alpha + IL4	0.0
93572_Secondary Th2_resting day 4-6 in IL-2	33.9	93781_HUVEC (Endothelial)_IL-11	0 2
93571_Secondary Tr1_resting day 4-6 in IL-2	40.1	93583_Lung Microvascular Endothelial Cells_none	0.1
93568_primary Th1_anti-CD28/anti-CD3	16.6	93584_Lung Microvascular Endothelial Cells_TNFa (4 ng/ml) and IL1b (1 ng/ml)	0.3
93569_primary Th2_anti-CD28/anti-CD3	48.3	92662_Microvascular Dermal endothelium_none	0.0
93570_primary Tr1_anti-CD28/anti-CD3	43.8	92663_Microsvasular Dermal endothelium_TNFa (4 ng/ml) and IL1b (1 ng/ml)	0.1
93565_primary Th1_resting dy 4-6 in IL-2	29.7	93773_Bronchial epithelium_TNFa (4 ng/ml) and IL1b (1 ng/ml) **	0.1
93566_primary Th2_resting dy 4-6 in IL-2	23.0	93347_Small Airway Epithelium_none	0.0
93567_primary Tr1_resting dy 4-6 in IL-2	79.0	93348_Small Airway Epithelium_TNFa (4 ng/ml) and IL1b (1 ng/ml)	0.2
93351_CD45RA CD4 lymphocyte_anti-CD28/anti-CD3	23.2	92668_Coronery Artery SMC_resting	0.0
93352_CD45RO CD4 lymphocyte_anti-CD28/anti-CD3	62.0	92669_Coronery Artery SMC_TNFa (4 ng/ml) and IL1b (1 ng/ml)	0.0
93251_CD8 Lymphocytes_anti-CD28/anti-CD3	37.4	93107_astrocytes_resting	0.0
93353_chronic CD8 Lymphocytes 2ry_resting dy 4-6 in IL-2	43.8	93108_astrocytes_TNFa (4 ng/ml) and IL1b (1 ng/ml)	0.0
93574_chronic CD8	29.3	92666_KU-812	0.9

Lymphocytes 2ry_activated CD3/CD28		(Basophil)_resting	
93354_CD4_none	14.2	92667_KU-812 (Basophil)_PMA/ionoycin	0.9
93252_Secondary Th1/Th2/Tr1_anti-CD95 CH11	40.9	93579_CCD1106 (Keratinocytes)_none	0.1
93103_LAK cells_resting	15.2	93580_CCD1106 (Keratinocytes)_TNFa and IFNg **	0.1
93788_LAK cells_IL-2	52.1	93791_Liver Cirrhosis	3.4
93787_LAK cells_IL-2+IL-12	23.5	93577_NCI-H292	0.3
93789_LAK cells_IL-2+IFN gamma	29.7	93358_NCI-H292_IL-4	0.0
93790_LAK cells_IL-2+ IL-18	37.1	93360_NCI-H292_IL-9	0.1
93104_LAK cells_PMA/ionomycin and IL-18	15.8	93359_NCI-H292_IL-13	0.1
93578_NK Cells IL-2_resting	100.0	93357_NCI-H292_IFN gamma	0 1
93109_Mixed Lymphocyte Reaction_Two Way MLR	27.5	93777_HPAEC_-	0 1
93110_Mixed Lymphocyte Reaction_Two Way MLR	27.4	93778_HPAEC_IL-1 beta/TNA alpha	0.0
93111_Mixed Lymphocyte Reaction_Two Way MLR	36.3	93254_Normal Human Lung Fibroblast_none	0 1
93112_Mononuclear Cells (PBMCs)_resting	20.6	93253_Normal Human Lung Fibroblast_TNFa (4 ng/ml) and IL-1b (1 ng/ml)	0.0
93113_Mononuclear Cells (PBMCs)_PWM	34.9	93257_Normal Human Lung Fibroblast_IL-4	0.0
93114_Mononuclear Cells (PBMCs)_PHA-L	40.3	93256_Normal Human Lung Fibroblast_IL-9	0.0
93249_Ramos (B cell)_none	2.6	93255_Normal Human Lung Fibroblast_IL-13	0.4
93250_Ramos (B cell)_ionomycin	3.0	93258_Normal Human Lung Fibroblast_IFN gamma	0.5
93349_B lymphocytes_PWM	27.0	93106_Dermal Fibroblasts CCD1070_resting	1.6
93350_B lymphoytes_CD40L and IL-4	6.8	93361_Dermal Fibroblasts CCD1070_TNF alpha 4 ng/ml	52.8
92665_EOL-1 (Eosinophil)_dbcAMP differentiated	11.0	93105_Dermal Fibroblasts CCD1070_IL-1 beta 1 ng/ml	0.2
93248_EOL-1 (Eosinophil)_dbcAMP/PMAion omycin	2.1	93772_dermal fibroblast_IFN gamma	0.3
93356_Dendritic Cells_none	2.3	93771_dermal fibroblast_IL-4	0.4
93355_Dendritic Cells_LPS 100 ng/ml	0.3	93892_Dermal fibroblasts_none	0.2
93775_Dendritic Cells_anti-CD40	0.2	99202_Neutrophils_TNFa+LPS	0.8
93774_Monocytes_resting	0.9	99203_Neutrophils_none	2.0
93776_Monocytes_LPS 50 ng/ml	1.9	735010_Colon_normal	2.8
93581_Macrophages_resting	1.1	735019_Lung_none	2.5
93582_Macrophages_LPS 100 ng/ml	0.9	64028-1_Thymus_none	19.3
93098_HUVEC (Endothelial)_none	0.0	64030-1_Kidney_none	9.5
93099_HUVEC (Endothelial)_starved	0.3		

Panel 2.1 Summary: Ag4004 The NOV13 - 87919652 gene is strongly expressed in normal lung tissues when compared to the associated lung tumor tissue in Panel 2.1, with highest expression in a normal lung tissue sample (CT = 29.6). Thus, based upon this profile, expression of this gene could be used as a marker to differentiate normal lung tissues from lung tumors. Furthermore, the 87919652 gene product may be useful as a protein therapeutic in the treatment of lung cancer, through the use of peptides, chimeric molecules and small molecule drugs.

Panel 4.1D Summary: Ag4004 The highest expression of the NOV13 - 87919652 gene is seen in NK cells (CT = 28.2 vs 29.1 to 33.1 in other activated T cells). Moderate expression of this gene is seen in other T cells irrespective of treatment. Besides lymphoid cells, the 87919652 gene is also highly expressed in dermal fibroblasts treated with TNFa. Therefore, modulation of the protein encoded for by the 87919652 gene could be important in immune-mediated diseases such as asthma, IBD, contact hypersensitivity, infection disease, allorejection and autoimmunity.

NOV14 - 87935554

Expression of gene 87935554 was assessed using the primer-probe set Ag3998, described in Table 25A. Results from RTQ-PCR runs are shown in Table 25B.

Table 25A. Probe Name Ag3998

Primers	Sequences	TM	Length	Start Position	SEQ ID NO
Forward	5'-CTGCCCTGCTACTTGCTCTAC-3'	59.3	21	215	143
Probe	FAM-5'-CACCATTGTCGTGGCTACATCATCCT-3'-TAMRA	69	26	242	144
Reverse	5'-AGGACCATCTTGAGCTTGGA-3'	59.8	20	278	145

Table 25B. Panel 4.1D

Tissue Name	Relative Expression(%) 4.1dx4tm6155f_ag3998_a2	Tissue Name	Relative Expression(%) 4.1dx4tm6155f_ag3998_a2
93768_Secondary Th1_anti-CD28/anti-CD3	0.2	93100_HUVEC (Endothelial)_IL-1b	0.7
93769_Secondary Th2_anti-CD28/anti-CD3	0.0	93779_HUVEC (Endothelial)_IFN gamma	0.7
93770_Secondary Tr1_anti-CD28/anti-CD3	0.2	93102_HUVEC (Endothelial)_TNF alpha + IFN gamma	0.4
93573_Secondary Th1_resting day 4-6 in IL-2	0.2	93101_HUVEC (Endothelial)_TNF alpha + IL4	0.3
93572_Secondary Th2_resting day 4-6 in IL-2	0.0	93781_HUVEC (Endothelial)_IL-11	1.5
93571_Secondary Tr1_resting day 4-6 in IL-2	0.0	93583_Lung Microvascular Endothelial Cells_none	27.4
93568_primary Th1_anti-CD28/anti-CD3	0.0	93584_Lung Microvascular Endothelial Cells_TNFa (4 ng/ml) and IL1b (1 ng/ml)	16.6

93569_primary Th2_anti-CD28/anti-CD3	0.1	92662_Microvascular Dermal endothelium_none	29.5
93570_primary Tr1_anti-CD28/anti-CD3	0.0	92663_Microsvasular Dermal endothelium_TNFa (4 ng/ml) and IL1b (1 ng/ml)	7.0
93565_primary Th1_resting dy 4-6 in IL-2	0.0	93773_Bronchial epithelium_TNFa (4 ng/ml) and IL1b (1 ng/ml) **	8.5
93566_primary Th2_resting dy 4-6 in IL-2	0.0	93347_Small Airway Epithelium_none	3.3
93567_primary Tr1_resting dy 4-6 in IL-2	0.0	93348_Small Airway Epithelium_TNFa (4 ng/ml) and IL1b (1 ng/ml)	16.2
93351_CD45RA CD4 lymphocyte_anti-CD28/anti-CD3	16.6	92668_Coronery Artery SMC_resting	0.5
93352_CD45RO CD4 lymphocyte_anti-CD28/anti-CD3	0.0	92669_Coronery Artery SMC_TNFa (4 ng/ml) and IL1b (1 ng/ml)	1.0
93251_CD8 Lymphocytes_anti-CD28/anti-CD3	0.0	93107_astrocytes_resting	16.9
93353_chronic CD8 Lymphocytes 2ry_resting dy 4-6 in IL-2	0.2	93108_astrocytes_TNFa (4 ng/ml) and IL1b (1 ng/ml)	12.7
93574_chronic CD8 Lymphocytes 2ry_activated CD3/CD28	0.0	92666_KU-812 (Basophil)_resting	0.0
93354_CD4_none	0.2	92667_KU-812 (Basophil)_PMA/ionoycin	0.1
93252_Secondary Th1/Th2/Tr1_anti-CD95 CH11	0.0	93579_CCD1106 (Keratinocytes)_none	10.8
93103_LAK cells_resting	18.0	93580_CCD1106 (Keratinocytes)_TNFa and IFNg **	7.9
93788_LAK cells_IL-2	0.1	93791_Liver Cirrhosis	17.7
93787_LAK cells_IL-2+IL-12	0.5	93577_NCI-H292	5.1
93789_LAK cells_IL-2+IFN gamma	0.7	93358_NCI-H292_IL-4	7.7
93790_LAK cells_IL-2+ IL-18	0.4	93360_NCI-H292_IL-9	8.0
93104_LAK cells_PMA/ionomycin and IL-18	10.5	93359_NCI-H292_IL-13	8.9
93578_NK Cells IL-2_resting	0.0	93357_NCI-H292_IFN gamma	4.3
93109_Mixed Lymphocyte Reaction_Two Way MLR	6.6	93777_HPAEC_-	1.3
93110_Mixed Lymphocyte Reaction_Two Way MLR	4.6	93778_HPAEC_IL-1 beta/TNA alpha	0.9
93111_Mixed Lymphocyte Reaction_Two Way MLR	1.7	93254_Normal Human Lung Fibroblast_none	11.1
93112_Mononuclear Cells (PBMCs)_resting	1.3	93253_Normal Human Lung Fibroblast_TNFa (4 ng/ml) and IL-1b (1 ng/ml)	6.7
93113_Mononuclear Cells (PBMCs)_PWM	0.7	93257_Normal Human Lung Fibroblast_IL-4	7.2
93114_Mononuclear Cells (PBMCs)_PHA-L	1.8	93256_Normal Human Lung Fibroblast_IL-9	13.3
93249_Ramos (B cell)_none	0.0	93255_Normal Human Lung Fibroblast_IL-13	7.6
93250_Ramos (B cell)_ionomycin	0.0	93258_Normal Human Lung Fibroblast_IFN gamma	7.6
93349_B lymphocytes_PWM	0.0	93106_Dermal Fibroblasts CCD1070_resting	26.2
93350_B lymphoytes_CD40L and IL-4	0.4	93361_Dermal Fibroblasts CCD1070_TNF alpha 4 ng/ml	25.1

92665_EOL-1 (Eosinophil)_dbcAMP differentiated	0.0	93105_Dermal Fibroblasts CCD1070_IL-1 beta 1 ng/ml	25.3
93248_EOL-1 (Eosinophil)_dbcAMP/PMAionomycin	0.0	93772_dermal fibroblast_IFN gamma	1.2
93356_Dendritic Cells_none	44.9	93771_dermal fibroblast_IL-4	0.9
93355_Dendritic Cells_LPS 100 ng/ml	55.4	93892_Dermal fibroblasts_none	2.4
93775_Dendritic Cells_anti-CD40	100.0	99202_Neutrophils_TNFa+LPS	0.4
93774_Monocytes_resting	5.3	99203_Neutrophils_none	0.4
93776_Monocytes_LPS 50 ng/ml	24.2	735010_Colon_normal	4.5
93581_Macrophages_resting	45.1	735019_Lung_none	7 5
93582_Macrophages_LPS 100 ng/ml	16.6	64028-1_Thymus_none	2.3
93098_HUVEC (Endothelial)_none	0.4	64030-1_Kidney_none	13.5
93099_HUVEC (Endothelial)_starved	0.9		

Panel 4.1D Summary: Ag3998 In lymphoid cells, the NOV14 - 87935554 gene is highly expressed in dendritic cells and in mature dendritic cells treated with anti-CD40 (CT = 26.3). Moderate to high expression of this gene is also found in monocytes and macrophages (independently of their activation), untreated LAK cells, activated naive T cells (but not memory T cells), fibroblasts (dermis and lung), and endothelial cells. This gene encodes a putative canalicular multispecific organic anion transporter, a member of the multidrug resistance-associated protein family; proteins in this family have been reported to play a widespread role in detoxification, drug resistance, and. due to their role in the export of glutathione disulfide by MRP1 and MRP2, in the defense against oxidative stress. See, Wijnholds *et al.*, Nat. Med. 3: 1275-1279, 1997. Therefore, regulation of the 87935554 gene product by small molecule therapeutics could be important in the treatment of inflammatory diseases and cancer.

The multidrug resistance-associated protein (MRP) mediates the cellular excretion of many drugs, glutathione S-conjugates (GS-X) of lipophilic xenobiotics and endogenous cysteinyl leukotrienes. Increased MRP levels in tumor cells can cause multidrug resistance (MDR) by decreasing the intracellular drug concentration. The physiological role or roles of MRP remain ill-defined, however. MRP-deficient mice have been generated by using embryonic stem cell technology. Mice homozygous for the mrp mutant allele, mrp-/-, are viable and fertile, but their response to an inflammatory stimulus is impaired. This defect is attribute to a decreased secretion of leukotriene C4 (LTC4) from leukotriene-synthesizing cells. Moreover, the mrp-/- mice are hypersensitive to the anticancer drug etoposide. The phenotype of mrp-/- mice is consistent with a role for MRP as the main LTC4-exporter in leukotriene-synthesizing cells, and as an important drug exporter in drug-sensitive cells. Results suggest that this ubiquitous GS-X

pump is dispensable in mice, making treatment of MDR with MRP-specific reversal agents potentially feasible. PMID: 9359705

NOV15a - 100399281

Expression of gene NOV15a - 100399281 was assessed using the primer-probe sets Ag391, Ag672, and Ag3999, described in Tables 26A, 26B, and 26C. Results from RTQ-PCR runs are shown in Table 26D and 26E.

Table 26A. Probe Name Ag391

Primers	Sequences	TM	Length	Start Position	SEQ ID NO
Forward	5'-GACGGTCACAGGTCCTCGAT-3'		20	629	146
Probe	TET-5'-TGCACGCGTAGCCACAAGACCG-3'-TAMRA		22	597	147
Reverse	5'-GGGAACGGCAACCAGAAAC-3'		19	573	148

Table 26B. Probe Name Ag672

Primers	Sequences	TM	Length	Start Position	SEQ ID NO
Forward	5'-CCAGATCCTTTCTCCTTGATCT-3'	58.8	22	172	149
Probe	TET-5'-CCAAACTTTCCAGATCTTTCCAAAGCTG-3'-TAMRA	68.5	28	195	150
Reverse	5'-TGACCTGGATATTTGGATTCTG-3'	58.9	22	234	151

Table 26C. Probe Name Ag3999

Primers	Sequences	TM	Length	Start Position	SEQ ID NO
Forward	5'-AACAGAATCGAGGACCTGTGA-3'	59.7	21	795	152
Probe	FAM-5'-CCAGCTTGCACCGGATTCCTGAT-3'-TAMRA	70.5	23	829	153
Reverse	5'-CCCTAACCAAGCTTCCTTTACA-3'	59.7	22	852	154

Table 26D. Panel 1

Tissue Name	Relative Expression(%) ltm408f	Tissue Name	Relative Expression(%) ltm408f
Endothelial cells	0.1	Kidney (fetal)	9.5
Endothelial cells (treated)	0.0	Renal ca. 786-0	3.9
Pancreas	12.1	Renal ca. A498	7.2
Pancreatic ca. CAPAN 2	1.6	Renal ca. RXF 393	16.0
Adipose	24.8	Renal ca. ACHN	1.0
Adrenal gland	4.2	Renal ca. UO-31	0.8
Thyroid	96.6	Renal ca. TK-10	20.2
Salavary gland	17.1	Liver	22.5
Pituitary gland	3.4	Liver (fetal)	0.4
Brain (fetal)	3.6	Liver ca. (hepatoblast) HepG2	2.2

Brain (whole)	6.7	Lung	6 2
Brain (amygdala)	5.0	Lung (fetal)	45.4
Brain (cerebellum)	17.7	Lung ca. (small cell) LX-1	4.8
Brain (hippocampus)	5.7	Lung ca. (small cell) NCI-H69	14.6
Brain (substantia nigra)	8.0	Lung ca. (s.cell var) SHP-77	5.7
Brain (thalamus)	14.4	Lung ca. (large cell)NCI-H460	3.6
Brain (hypothalamus)	9.5	Lung ca. (non-sm. cell) A549	3.1
Spinal cord	11.9	Lung ca. (non-s.cell) NCI-H23	1.4
CNS ca. (glio/astro) U87-MG	2.8	Lung ca (non-s.cell) HOP-62	0 6
CNS ca (glio/astro) U-118-MG	9.3	Lung ca. (non-s.cl) NCI-H522	0.4
CNS ca. (astro) SW1783	0.7	Lung ca. (squam.) SW 900	34.6
CNS ca.* (neuro; met) SK-N-AS	0.9	Lung ca. (squam.) NCI-H596	35.4
CNS ca. (astro) SF-539	1.1	Mammary gland	100.0
CNS ca. (astro) SNB-75	3.7	Breast ca.* (pl. effusion) MCF-7	4.3
CNS ca. (glio) SNB-19	1.8	Breast ca.* (pl.ef) MDA-MB-231	1.5
CNS ca. (glio) U251	1.4	Breast ca.* (pl. effusion) T47D	8.3
CNS ca. (glio) SF-295	0.6	Breast ca. BT-549	3.7
Heart	5.8	Breast ca. MDA-N	0.6
Skeletal muscle	4.1	Ovary	2.8
Bone marrow	3.3	Ovarian ca. OVCAR-3	2.3
Thymus	11.0	Ovarian ca. OVCAR-4	0.9
Spleen	15.1	Ovarian ca. OVCAR-5	4.4
Lymph node	5.7	Ovarian ca. OVCAR-8	3.1
Colon (ascending)	5.4	Ovarian ca. IGROV-1	1.6
Stomach	13.7	Ovarian ca.* (ascites) SK-OV-3	2.2
Small intestine	9.4	Uterus	11.7
Colon ca. SW480	7.0	Placenta	95.9
Colon ca * (SW480 met)SW620	1.1	Prostate	6.2
Colon ca. HT29	1.6	Prostate ca.* (bone met)PC-3	0.3
Colon ca. HCT-116	3.3	Testis	5.7
Colon ca. CaCo-2	2.9	Melanoma Hs688(A).T	0.6
Colon ca. HCT-15	2.7	Melanoma* (met) Hs688(B).T	0.3
Colon ca. HCC-2998	2.3	Melanoma UACC-62	1.4
Gastric ca.* (liver met) NCI-N87	5.7	Melanoma M14	1.4
Bladder	10.4	Melanoma LOX IMVI	1.6
Trachea	8.8	Melanoma* (met) SK-MEL-5	23.2
Kidney	13.7	Melanoma SK-MEL-28	18.8

Table 26E. Panel 1.1

Tissue Name	Relative Expression(%) 1.1tm798t_ag672	Tissue Name	Relative Expression(%) 1.1tm798t_ag672
Adipose	3.2	Renal ca. TK-10	36.1
Adrenal gland	3.8	Renal ca. UO-31	0.1
Bladder	24.3	Renal ca. RXF 393	11.6
Brain (amygdala)	0.2	Liver	5.0
Brain (cerebellum)	3.7	Liver (fetal)	0.0
Brain (hippocampus)	3.1	Liver ca. (hepatoblast) HepG2	0.6
Brain (substantia nigra)	14.4	Lung	6.5
Brain (thalamus)	10.4	Lung (fetal)	57.4
Cerebral Cortex	3.2	Lung ca (non-s.cell) HOP-62	0.6
Brain (fetal)	1.3	Lung ca. (large cell)NCI-H460	4.9
Brain (whole)	1.5	Lung ca. (non-s.cell) NCI-H23	0.1
CNS ca (glio/astro) U-118-MG	15.3	Lung ca. (non-s.cl) NCI-H522	0.0
CNS ca. (astro) SF-539	0.0	Lung ca. (non-sm. cell) A549	2.5
CNS ca. (astro) SNB-75	0.6	Lung ca. (s.cell var.) SHP-77	0.0
CNS ca. (astro) SW1783	0.0	Lung ca. (small cell) LX-1	11.6
CNS ca. (glio) U251	1.0	Lung ca. (small cell) NCI-H69	17.7
CNS ca. (glio) SF-295	0.0	Lung ca. (squam.) SW 900	29.3
CNS ca. (glio) SNB-19	0.8	Lung ca. (squam.) NCI-H596	63.3
CNS ca. (glio/astro) U87-MG	3.7	Lymph node	3.4
CNS ca.* (neuro; met) SK-N-AS	0.0	Spleen	3.8
Mammary gland	43.5	Thymus	2.5
Breast ca. BT-549	0.8	Ovary	0.8
Breast ca. MDA-N	0.1	Ovarian ca. IGROV-1	5.0
Breast ca.* (pl. effusion) T47D	9.6	Ovarian ca. OVCAR-3	6.3
Breast ca.* (pl. effusion) MCF-7	2.7	Ovarian ca. OVCAR-4	0.0
Breast ca.* (pl.ef) MDA-MB-231	1.2	Ovarian ca. OVCAR-5	6.2
Small intestine	6.2	Ovarian ca. OVCAR-8	1.8
Colorectal	0.1	Ovarian ca.* (ascites) SK-OV-3	2.7
Colon ca. HT29	0.0	Pancreas	54.0
Colon ca. CaCo-2	2.1	Pancreatic ca. CAPAN 2	0.1
Colon ca. HCT-15	1.7	Pituitary gland	14.0
Colon ca. HCT-116	1.7	Placenta	100.0
Colon ca. HCC-2998	2.2	Prostate	1.0
Colon ca. SW480	19.1	Prostate ca.* (bone met)PC-3	0.0
Colon ca.* (SW480 met)SW620	0.8	Salavary gland	42.3

Stomach	6.7	Trachea	23.8
Gastric ca.* (liver met) NCI-N87	9.5	Spinal cord	6.7
Heart	23.0	Testis	0.0
Fetal Skeletal	1.6	Thyroid	78.5
Skeletal muscle	13.1	Uterus	1.6
Endothelial cells	0.2	Melanoma M14	0.0
Heart (fetal)	4.2	Melanoma LOX IMVI	0.0
Kidney	6.4	Melanoma UACC-62	0.0
Kidney (fetal)	5.0	Melanoma SK-MEL-28	65.5
Renal ca. 786-0	2.0	Melanoma* (met) SK-MEL-5	37.1
Renal ca. A498	13.2	Melanoma Hs688(A).T	0.0
Renal ca. ACHN	0.1	Melanoma* (met) Hs688(B).T	0.0

Panel 1 Summary: Ag391 Two experiments were performed using the same probe/primer set; results from one of the replicate runs were discarded because the results were artifactual (data not shown). The NOV15a - 100399281 gene is moderately to highly expressed across the majority of samples on this panel. However, expression is highest in mammary gland (CT = 26), placenta (CT = 26.1), and thyroid (CT = 26.1). Therefore, the 100399281 gene might be useful as a marker to distinguish these tissues. In addition, the observed expression in mammary gland and placenta suggests a potential role for the 100399281 gene product in pregnancy. Interestingly, expression of this gene is much lower in 5/5 breast cancer cell lines when compared to normal breast. This suggests that replacement of the 100399281 gene product using protein therapeutics, peptides or gene therapy would be valuable in the treatment of breast cancer.

In addition, the NOV15a - 100399281 gene is expressed throughout the CNS with moderate expression detected in amygdala, cerebellum, hippocampus, substantia nigra, thalamus, hypothalamus and spinal cord. Expression of this gene is decreased in CNS cancer cell lines relative to normal brain tissues. The secreted protein encoded for by the 100399281 gene contains homology to thrombospondin, suggesting it may play a role in inhibiting angiogenesis. Therefore, treatment with the 100399281 protein, or in vivo modulation of the gene or the protein product may therefore be of use in slowing the growth/ inhibiting CNS tumors. Selective removal of this protein via synthetic antibodies may help to increase vascularization in CNS tissue undergoing repair/regeneration.

Among the metabolically relevant tissues, the NOV15a - 100399281 gene is expressed at high levels in thyroid and at more moderate levels in pancreas, adrenal gland, pituitary gland, heart, and skeletal muscle. Therefore, this gene product may have utility as a drug treatment for any or all diseases of the thyroid gland as well as other metabolic and neuroendocrine diseases.

Interestingly, this gene is more highly expressed in adult liver (CT = 28.2) than in fetal liver (CT = 33.8), suggesting that the 100399281 gene would be a useful marker for differentiating between the adult and fetal liver. Please note that the adipose sample on this panel is contaminated with genomic DNA and, therefore, expression in this tissue cannot be analyzed.

Panel 1.1 Summary: Ag672 The results obtained in this experiment are comparable to what is observed in Panel 1. Expression of the NOV15a - 100399281 gene is primarily associated with normal tissues on this panel. Highest expression is seen in placenta (CT = 25), thyroid (CT = 25.2), pancreas (CT = 25.7), and mammary gland (CT = 26). Therefore, the 100399281 gene might be useful as a marker to distinguish these tissues. In addition, the observed expression in mammary gland and placenta suggests a potential role for the 100399281 gene product in pregnancy. Interestingly, expression of this gene is much lower in 5/5 breast cancer cell lines when compared to normal breast. This suggests that replacement of the 100399281 gene product using protein therapeutics, peptides or gene therapy would be valuable in the treatment of breast cancer.

In addition, the 100399281 gene is expressed throughout the CNS with low to moderate expression detected in amygdala, cerebellum, hippocampus, substantia nigra, thalamus and cerebral cortex. Expression of this gene is decreased in CNS cancer cell lines relative to normal brain tissues. The secreted protein encoded for by the 100399281 gene contains homology to thrombospondin, suggesting it may play a role in inhibiting angiogenesis. Therefore, treatment with the 100399281 protein, or in vivo modulation of the gene or the protein product may therefore be of use in slowing the growth/ inhibiting CNS tumors. Selective removal of this protein via synthetic antibodies may help to increase vascularization in CNS tissue undergoing repair/regeneration.

Among the metabolically relevant tissues, the 100399281 gene is expressed at high levels in thyroid and pancreas and at more moderate levels in adrenal gland, pituitary gland, heart, and skeletal muscle. Therefore, this gene product may have utility as a drug treatment for any or all diseases of the thyroid gland and pancreas as well as other metabolic and neuroendocrine diseases. Interestingly, this gene is more highly expressed in adult liver (CT = 29) than in fetal liver (CT = 40), suggesting that the 100399281 gene would be a useful marker for differentiating between the adult and fetal liver. Please note that the adipose sample on this panel is contaminated with genomic DNA and, therefore, expression in this tissue cannot be analyzed.

Panel 2.1 Summary: Ag3999 Expression of the NOV15a - 100399281 gene is low/undetectable (CT values > 35) across the samples on this panel (data not shown).

Panel 4.1D Summary: Ag3999 Expression of the NOV15a - 100399281 gene is low/undetectable (CT values > 35) across the samples on this panel (data not shown).

NOV16a - 101330077

Expression of gene NOV16a - 101330077 was assessed using the primer-probe set Ag3996, described in Table 27A. Results from RTQ-PCR run are shown in Table 27B.

Table 27A. Probe Name Ag3996

Primers	Sequences	TM	Length	Start Position	SEQ ID NO
Forward	5'-GAGTGGGCTACACCAATCAG-3'	58.2	20	411	155
Probe	FAM-5'-AGCGGCGCTAACGTGACTGACTAACT-3'-TAMRA	69	26	437	156
Reverse	5'-CCCTCTCAGGGAGATTGAGA-3'	59.3	20	476	157

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Table 27B. Panel 4.1D

Tissue Name	Relative Expression(%)	
	4.1dtm6144f_ag3996	4.1dx4tm6155f_ag3996_a1
93768_Secondary Th1_anti-CD28/anti-CD3	1.3	0.0
93769_Secondary Th2_anti-CD28/anti-CD3	1.9	2.7
93770_Secondary Tr1_anti-CD28/anti-CD3	0.0	0.0
93573_Secondary Th1_resting day 4-6 in IL-2	0.0	0.0
93572_Secondary Th2_resting day 4-6 in IL-2	0.7	0.0
93571_Secondary Tr1_resting day 4-6 in IL-2	0.0	1.1
93568_primary Th1_anti-CD28/anti-CD3	0.0	0.0
93569_primary Th2_anti-CD28/anti-CD3	0.0	0.0
93570_primary Tr1_anti-CD28/anti-CD3	0.8	0.0
93565_primary Th1_resting dy 4-6 in IL-2	0.4	0.0
93566_primary Th2_resting dy 4-6 in IL-2	0.0	0.0
93567_primary Tr1_resting dy 4-6 in IL-2	0.4	0.0
93351_CD45RA CD4 lymphocyte_anti-CD28/anti-CD3	0.0	0.0
93352_CD45RO CD4 lymphocyte_anti-CD28/anti-CD3	0.6	0.0
93251_CD8 Lymphocytes_anti-CD28/anti-CD3	0.0	0.0
93353_chronic CD8 Lymphocytes 2ry_resting dy 4-6 in IL-2	0.0	0.0
93574_chronic CD8 Lymphocytes 2ry_activated CD3/CD28	0.0	0.0
93354_CD4_none	0.0	0.0
93252_Secondary Th1/Th2/Tr1_anti-CD95 CH11	0.8	1.3
93103_LAK cells_resting	0.0	0.0
93788_LAK cells_IL-2	0.0	0.0
93787_LAK cells_IL-2+IL-12	0.0	0.0
93789_LAK cells_IL-2+IFN gamma	0.0	0.0
93790_LAK cells_IL-2+ IL-18	0.5	0.0
93104_LAK cells_PMA/ionomycin and IL-18	0.0	0.0
93578_NK Cells IL-2_resting	0.5	1.6
93109_Mixed Lymphocyte Reaction_Two Way MLR	0.0	0.0
93110_Mixed Lymphocyte Reaction_Two Way MLR	0.0	0.0
93111_Mixed Lymphocyte Reaction_Two Way MLR	0.0	0.0

93112_Mononuclear Cells (PBMCs)_resting	0.0	1 2
93113_Mononuclear Cells (PBMCs)_PWM	0.4	1 2
93114_Mononuclear Cells (PBMCs)_PHA-L	0.0	0.0
93249_Ramos (B cell)_none	0.0	0.0
93250_Ramos (B cell)_ionomycin	0.0	0.0
93349_B lymphocytes_PWM	1.4	0.0
93350_B lymphoytes_CD40L and IL-4	0.0	0.0
92665_EOL-1 (Eosinophil)_dbcAMP differentiated	0.0	0.0
93248_EOL-1 (Eosinophil)_dbcAMP/PMAionomycin	0.0	0 0
93356_Dendritic Cells_none	0.0	0.0
93355_Dendritic Cells_LPS 100 ng/ml	0.0	0.0
93775_Dendritic Cells_anti-CD40	0.0	0.0
93774_Monocytes_resting	0.0	0.0
93776_Monocytes_LPS 50 ng/ml	0.0	0.0
93581_Macrophages_resting	0.0	0.0
93582_Macrophages_LPS 100 ng/ml	1.8	2.2
93098_HUVEC (Endothelial)_none	0.0	0.0
93099_HUVEC (Endothelial)_starved	0.0	0.0
93100_HUVEC (Endothelial)_IL-1b	0.6	0.0
93779_HUVEC (Endothelial)_IFN gamma	0.0	0.0
93102_HUVEC (Endothelial)_TNF alpha + IFN gamma	0.0	0.0
93101_HUVEC (Endothelial)_TNF alpha + IL4	0.0	0.0
93781_HUVEC (Endothelial)_IL-11	0.0	0.0
93583_Lung Microvascular Endothelial Cells_none	0.7	3.1
93584_Lung Microvascular Endothelial Cells_TNFa (4 ng/ml) and IL1b (1 ng/ml)	0.0	0.0
92662_Microvascular Dermal endothelium_none	0.0	0.0
92663_Microsvasular Dermal endothelium_TNFa (4 ng/ml) and IL1b (1 ng/ml)	0.0	0.0
93773_Bronchial epithelium_TNFa (4 ng/ml) and IL1b (1 ng/ml) **	0.0	0.0
93347_Small Airway Epithelium_none	0.0	0.0
93348_Small Airway Epithelium_TNFa (4 ng/ml) and IL1b (1 ng/ml)	0.0	0.0
92668_Coronery Artery SMC_resting	0.4	1.1
92669_Coronery Artery SMC_TNFa (4 ng/ml) and IL1b (1 ng/ml)	0.0	0.0
93107_astrocytes_resting	0.0	0.4
93108_astrocytes_TNFa (4 ng/ml) and IL1b (1 ng/ml)	0.2	0.0
92666_KU-812 (Basophil)_resting	0.0	0.0
92667_KU-812 (Basophil)_PMA/ionoycin	0.0	0.0
93579_CCD1106 (Keratinocytes)_none	0.3	0.0
93580_CCD1106 (Keratinocytes)_TNFa and IFNg **	0.0	5.0
93791_Liver Cirrhosis	0.0	0.0
93577_NCI-H292	0.0	0.0
93358_NCI-H292_IL-4	0.0	0.7
93360_NCI-H292_IL-9	1.0	0.8
93359_NCI-H292_IL-13	0.0	0.0
93357_NCI-H292_IFN gamma	0.0	0.0

93777_HPAEC_-	0.0	0.0
93778_HPAEC_IL-1 beta/TNA alpha	0.0	0.0
93254_Normal Human Lung Fibroblast_none	0.0	0.0
93253_Normal Human Lung Fibroblast_TNFa (4 ng/ml) and IL-1b (1 ng/ml)	0.0	0.0
93257_Normal Human Lung Fibroblast_IL-4	0.0	0.0
93256_Normal Human Lung Fibroblast_IL-9	0.0	0.0
93255_Normal Human Lung Fibroblast_IL-13	2.3	0.0
93258_Normal Human Lung Fibroblast_IFN gamma	2.9	0.0
93106_Dermal Fibroblasts CCD1070_resting	0.0	2.4
93361_Dermal Fibroblasts CCD1070_TNF alpha 4 ng/ml	3.4	0.0
93105_Dermal Fibroblasts CCD1070_IL-1 beta 1 ng/ml	1.7	0.0
93772_dermal fibroblast_IFN gamma	0.0	2.2
93771_dermal fibroblast_IL-4	0.9	0.0
93892_Dermal fibroblasts_none	1.9	0.0
99202_Neutrophils_TNFa+LPS	0.7	7.0
99203_Neutrophils_none	0.0	6.9
735010_Colon_normal	2.6	0.0
735019_Lung_none	11.8	6.0
64028-1_Thymus_none	27.5	19.6
64030-1_Kidney_none	100.0	100.0

Panel 2.1 Summary: Ag3996 Expression of the NOV16a - 101330077 gene is low/undetectable (CT values > 35) across the samples on this panel (data not shown).

Panel 4.1D Summary: Ag3996 Results from two experiments using the same probe/primer set are in fair agreement. Low but significant expression of the NOV16a - 101330077 gene is detected only in kidney and thymus. Therefore, the 101330077 transcript, the protein encoded for by this gene or antibodies designed against this gene product could be used to identify kidney and thymus tissue.

Example 3. SNP analysis of NOVX clones

SeqCallingTM Technology: cDNA was derived from various human samples representing multiple tissue types, normal and diseased states, physiological states, and developmental states from different donors. Samples were obtained as whole tissue, cell lines, primary cells or tissue cultured primary cells and cell lines. Cells and cell lines may have been treated with biological or chemical agents that regulate gene expression for example, growth factors, chemokines, steroids. The cDNA thus derived was then sequenced using CuraGen's proprietary SeqCalling technology. Sequence traces were evaluated manually and edited for corrections if appropriate. cDNA sequences from all samples were assembled with themselves and with public ESTs using bioinformatics programs to generate CuraGen's human SeqCalling database of SeqCalling assemblies. Each assembly contains one or more overlapping cDNA

sequences derived from one or more human samples. Fragments and ESTs were included as components for an assembly when the extent of identity with another component of the assembly was at least 95% over 50 bp. Each assembly can represent a gene and/or its variants such as splice forms and/or single nucleotide polymorphisms (SNPs) and their combinations.

Variant sequences are included in this application. A variant sequence can include a single nucleotide polymorphism (SNP). A SNP can, in some instances, be referred to as a "cSNP" to denote that the nucleotide sequence containing the SNP originates as a cDNA. A SNP can arise in several ways. For example, a SNP may be due to a substitution of one nucleotide for another at the polymorphic site. Such a substitution can be either a transition or a transversion. A SNP can also arise from a deletion of a nucleotide or an insertion of a nucleotide, relative to a reference allele. In this case, the polymorphic site is a site at which one allele bears a gap with respect to a particular nucleotide in another allele. SNPs occurring within genes may result in an alteration of the amino acid encoded by the gene at the position of the SNP. Intragenic SNPs may also be silent, however, in the case that a codon including a SNP encodes the same amino acid as a result of the redundancy of the genetic code. SNPs occurring outside the region of a gene, or in an intron within a gene, do not result in changes in any amino acid sequence of a protein but may result in altered regulation of the expression pattern for example, alteration in temporal expression, physiological response regulation, cell type expression regulation, intensity of expression, stability of transcribed message.

Method of novel SNP Identification: SNPs are identified by analyzing sequence assemblies using CuraGen's proprietary SNPTool algorithm. SNPTool identifies variation in assemblies with the following criteria: SNPs are not analyzed within 10 base pairs on both ends of an alignment; Window size (number of bases in a view) is 10; The allowed number of mismatches in a window is 2; Minimum SNP base quality (PHRED score) is 23; Minimum number of changes to score an SNP is 2/assembly position. SNPTool analyzes the assembly and displays SNP positions, associated individual variant sequences in the assembly, the depth of the assembly at that given position, the putative assembly allele frequency, and the SNP sequence variation. Sequence traces are then selected and brought into view for manual validation. The consensus assembly sequence is imported into CuraTools along with variant sequence changes to identify potential amino acid changes resulting from the SNP sequence variation. Comprehensive SNP data analysis is then exported into the SNPCalling database.

Method of novel SNP Confirmation: SNPs are confirmed employing a validated method know as Pyrosequencing (Pyrosequencing, Westborough, MA). Detailed protocols for Pyrosequencing can be found in: Alderborn *et al*. Determination of Single Nucleotide Polymorphisms by Real-time Pyrophosphate DNA Sequencing. (2000). *Genome Research.* 10,

Issue 8, August. 1249-1265. In brief. Pyrosequencing is a real time primer extension process of genotyping. This protocol takes double-stranded. biotinylated PCR products from genomic DNA samples and binds them to streptavidin beads. These beads are then denatured producing single stranded bound DNA. SNPs are characterized utilizing a technique based on an indirect bioluminometric assay of pyrophosphate (PPi) that is released from each dNTP upon DNA chain elongation. Following Klenow polymerase-mediated base incorporation, PPi is released and used as a substrate, together with adenosine 5'-phosphosulfate (APS), for ATP sulfurylase, which results in the formation of ATP. Subsequently, the ATP accomplishes the conversion of luciferin to its oxi-derivative by the action of luciferase. The ensuing light output becomes proportional to the number of added bases, up to about four bases. To allow processivity of the method dNTP excess is degraded by apyrase, which is also present in the starting reaction mixture, so that only dNTPs are added to the template during the sequencing. The process has been fully automated and adapted to a 96-well format, which allows rapid screening of large SNP panels. The DNA and protein sequences for the novel single nucleotide polymorphic variants are reported. Variants are reported individually but any combination of all or a select subset of variants are also included. In addition, the positions of the variant bases and the variant amino acid residues are underlined.

Results

Variants are reported individually but any combination of all or a select subset of variants are also included as contemplated NOVX embodiments of the invention.

NOV2 SNP data:

In the following positions os SEQ ID NO:9, one or more consensus positions (Cons. Pos.) of the nucleotide sequence have been identified as SNPs. "Depth" rerepresents the number of clones covering the region of the SNP. The Putative Allele Frequency (Putative Allele Freq.) is the fraction of all the clones containing the SNP. A dash ("-"), when shown, means that a base is not present. The sign ">" means "is changed to".

Cons.Pos.: 7216 Depth: 31 Change: C > T ; Cons.Pos.: 7118 Depth: 31 Change: C > T ; Cons.Pos.: 7266 Depth: 31 Change: T > A ; Cons.Pos.: 7328 Depth: 31 Change: C > T ; Cons.Pos.: 7355 Depth: 35 Change: C > T ; Cons.Pos.: 7365 Depth: 38 Change: C > T ; Cons.Pos.: 7368 Depth: 38 Change: C > T ; Cons.Pos.: 7451 Depth: 27 Change: G > A.

NOV3 SNP data:

A NOV3 variant cDNA, CG56383-01, was cloned that extended from nucleotide 1938 to 3129 of SEQ ID NO:5. SNP variants found in CG56383-01 are shown in Table 28. Two of the

SNPs are in the coding sequence of NOV3, with one change from T to C at nucleotide position 2089, and the other change from T to A at nucleotid position 2630. Two additional SNPs are in the 3' non-coding region, with two nucleotides (both Ts) at nucleotide position 3019-3020 deleted when compared to SEQ ID NO:10. The NOV3 sense strand (SEQ ID NO:10) and encoded polypeptide (SEQ ID NO:611) are used in Table 28 as the reference sequences to determine the base positions of the cSNPs and coding variants.

Table 28. cSNP and Coding Variants for NOV3

NT Position of cSNP	Wild Type NT	Variant NT	Amino Acid position	Amino Acid Change
2089	T	C	N/A	None
2630	T	A	214	Ser > Thr
3019	T	deletion	N/A	N/A
3020	T	deletion	N/A	N/A

NOV4 SNP data:

One or more consensus positions (Cons. Pos.) of the nucleotide sequence have been identified as SNPs as shown in Table 29. "Depth" represents the number of clones covering Cons.Pos.: 75 Depth: 18 Change: T > C ; Cons.Pos.: 517 Depth: 20 Change: T > C ; the region of the SNP. The Putative Allele Frequency (Putative Allele Freq.) is the fraction of all the clones containing the SNP. A dash ("-"), when shown, means that a base is not present. The sign ">" means "is changed to". Cons.Pos.: 75 Depth: 18 Change: T > C ; Cons.Pos.: 517 Depth: 20 Change: T > C.

NOV4 has two SNP variants, whose variant positions for their nucleotide and amino acid sequences are numbered according to SEQ ID NOs:18 and 19, respectively. The nucleotide sequences of these NOV4 variants differ as shown in Table 29.

Table 29. cSNP and Coding Variants for NOV4

NT Position of cSNP	Wild Type NT	Variant NT	Amino Acid position	Amino Acid Change
718	A	G	179	I > V
1134	A	G	N/A	None

NOV5 SNP data:

NOV5 has ten SNP variants, whose variant positions for their nucleotide and amino acid sequences are numbered according to SEQ ID NOs:26 and 27, respectively. The nucleotide sequences of these NOV5 variants differ as shown in Table 30.

Table 30. cSNP and Coding Variants for NOV5				
NT Position of cSNP	Wild Type NT	Variant NT	Amino Acid position	Amino Acid Change
172	A	G	36	E > K
203	T	C	N/A	None
273	T	C	70	S > P
283	G	A	73	G > E
287	C	T	N/A	None
381	G	T	106	D > Y
424	C	T	120	A > V
460	A	G	132	Q > R
504	G	A	147	E > K
559	C	T	165	S > F

NOV7 SNP data:

NOV7 has four SNP variants, whose variant positions for their nucleotide and amino acid sequences are numbered according to SEQ ID NOs: 43 and 43, respectively. The nucleotide sequences of these NOV7 variants differ as shown in Table 31.

Table 31. cSNP and Coding Variants for NOV7				
NT Position of cSNP	Wild Type NT	Variant NT	Amino Acid position	Amino Acid Change
187	T	C	N/A	None
222	T	C	16	V > A
229	A	G	N/A	None
377	A	G	68	N > D

NOV8 SNP data:

NOV8 has two SNP variants. whose variant positions for their nucleotide and amino acid sequences are numbered according to SEQ ID NOs:50 and 51, respectively. The nucleotide sequences of these NOV8 variants differ as shown in Table 32.

Table 32. cSNP and Coding Variants for NOV8				
NT Position of cSNP	Wild Type NT	Variant NT	Amino Acid position	Amino Acid Change
2060	G	A	N/A	None
2127	T	C	73	F > L

NOV9 SNP data:

NOV9 has three SNP variants, whose variant positions for their nucleotide and amino acid sequences are numbered according to SEQ ID NOs:52 and 53, respectively. The nucleotide sequences of these NOV9 variants differ as shown in Table 33.

Table 33. cSNP and Coding Variants for NOV9

NT Position of cSNP	Wild Type NT	Variant NT	Amino Acid position	Amino Acid Change
206	C	T	69	A > V
615	G	T	N/A	None
649	A	G	217	M > V

NOV10 SNP data:

The novel variants for the DNA and protein sequence for the novel hypothetical 22.2 kDa protein SLR0305-like / Type IIIb plasma membrane-like gene are reported here as variant Acc. No. 100340173. Variants are reported individually but any combination of all or a select subset of variants are also included.

NOV10 has four SNP variants, whose variant positions for their nucleotide and amino acid sequences are numbered according to SEQ ID NOs:60 and 61, respectively. The nucleotide sequences of these NOV10 variants differ as shown in Table 34.

Table 34. cSNP and Coding Variants for NOV10

NT Position of cSNP	Wild Type NT	Variant NT	Amino Acid position	Amino Acid Change
542	C	T	59	T > I
643	C	T	93	L > F
645	T	C	N/A	None
667	A	G	101	T > A

NOV12 SNP data:

NOV12 has one SNP variant, whose variant position for its nucleotide and amino acid sequences is numbered according to SEQ ID NOs:72 and 73, respectively. The nucleotide sequence of the NOV12 variant differs as shown in Table 35.

Table 35. cSNP and Coding Variants for NOV12

NT Position of cSNP	Wild Type NT	Variant NT	Amino Acid position	Amino Acid Change
2048	A	G	87	H > R

NOV15 SNPs and cSNPs:

One or more consensus positions (Cons. Pos.) of the nucleotide sequence have been identified as SNPs. "Depth" represents the number of clones covering the region of the SNP. The Putative Allele Frequency (Putative Allele Freq.) is the fraction of all the clones containing the SNP. A dash ("-"), when shown, means that a base is not present. The sign ">" means "is changed to".

Cons.Pos.: 648 Depth: 6 Change: - > A Putative Allele Freq.: 0.333 AA translation view (alpha) Fragment Listing: -> 14691381 2(+,i,119650936) Fpos: 137 -> 147572388 (+,i,119650936) Fpos: 172 Multi-Trace View

OTHER EMBODIMENTS

Although particular embodiments have been disclosed herein in detail, this has been done by way of example for purposes of illustration only, and is not intended to be limiting with respect to the scope of the appended claims, which follow. In particular, it is contemplated by the inventors that various substitutions, alterations, and modifications may be made to the invention without departing from the spirit and scope of the invention as defined by the claims. The choice of nucleic acid starting material, clone of interest, or library type is believed to be a matter of routine for a person of ordinary skill in the art with knowledge of the embodiments described herein. Other aspects, advantages, and modifications considered to be within the scope of the following claims.